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PULMONARY ADENOMATOSIS OF MAN

A Review of the Literature and a Report of Nine Cases

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NEW ORLEANS

THE pulmonary adenomatoses, or so-called alveolar cell tumors of the human lung, constitute a unique histologic group among the primary pulmonary neoplasms. There is general agreement that the majority of primary tumors of the lung arise from the bronchial respiratory epithelium. There is little unanimity of opinion, however, regarding the histogenesis of the adenomatoses, although most investigators concur that the site of origin is independent of the bronchi.

There are few conditions which offer as great a challenge to investigators as pulmonary adenomatosis, for whether or not it is a true neoplasm is still a matter of controversy. The etiologic concept is obscure and is complicated by conflicting claims of proponents of diverse theories. A unique opportunity to gain an insight into these problems was made possible by studying the relatively large series at the Army Institute of Pathology. Approximately 900 pulmonary neoplasms were screened to discover all cases of pulmonary adenomatosis which might have been filed under a designation other than those usually employed. Nine cases were found which satisfactorily fulfilled the criteria established for the purposes of this study.

It is the object of this paper (1) to review cases not included in those collected by Neubuerger and Geever [1] in 1942, (2) to present and discuss 9 cases from the files of the Army Institute of Pathology, (3) to examine theories concerning histogenesis and cause and (4) to discuss the question of whether pulmonary adenomatosis is related to carcinoma of the lung.

CRITERIA

It is unfortunate that precise criteria have not been fixed to limit the use of the term "pulmonary adenomatosis." Therefore, it is proposed that it be reserved for tumors which fulfil the following criteria: (1) alveolar cellular proliferation, characterized by the appearance of tall columnar mucus-producing cells; (2) absence of an intrinsic tumor of the bronchial tree, and (3) absence of primary adenocarcinoma of any

From the Army Institute of Pathology, Washington, D. C.

1. Neubuerger, K. T., and Geever, E. F., Arch. Path. **33**:551, 1942.

other part of the body. Those tumors which had involved regional lymph nodes or given rise to distant metastases but which otherwise conformed to the specified pattern were also included and were regarded as cancerous variations of pulmonary adenomatosis. Investigation has disclosed that adenomatosis has often been classified as an unusual example of bronchogenic carcinoma; therefore, it is not improbable that neoplasms thus diagnosed may have been included in material from which statistics pertaining to adenocarcinoma of the lung have been derived.

NOMENCLATURE

Some concept of the difficulties inherent in searching the literature for accounts of these tumors can be gained by considering the diversity of names selected. Judging from the names, one perceives that some authors considered this neoplasm benign; others, cancerous. There is still another group who have been noncommittal regarding origin or prognosis and have chosen unprejudicial designations. Among the many appellations are: "adenoma-like tumor," [2] "pulmonary adematosis," [3] "primary multiple alveolar cell tumor," [4] "papillary gelatinous adenocarcinoma." [5] "alveolar cell cancer," [6] "multicentric alveolar cell carcinoma," [7] "carcinomatoides alveogenica multicentrica," [8] "diffuse primary alveolar epithelial carcinoma," [9] "mucocellular papillary adenocarcinoma," [10] "malignant adenomatosis," [11] "carcinosis," [12] and "diffuse epithelial hyperplasia." [13] Recently "alveolar cell tumor" has become a popular choice,[14] but unfortunately it is often misinterpreted to mean that the tumor is derived from "alveolar lining cells" or has the morphologic configuration of alveoli, although the authors did not intend either implication.

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2. Helly, K.: Ztschr. f. Heilk. **28**:105, 1907.
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4. Neubuerger, K. T.: J. Thoracic Surg. **10**:557, 1941.
5. Briese: Frankfurt Ztschr. f. Path. **23**:48, 1920.
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7. Reuss, H.: Ueber zwei Fälle multicentrisch entstandener Lungenkrebse, Inaug. Dissert. Hamburg, 1934.
8. Casilli, A. R., and White, H. J.: Am. J. Clin. Path. **10**:623, 1940.
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11. Dacie, J. V., and Hoyle, C.: Brit. J. Tuberc. **36**:158, 1942.
12. Bonne, C.: Am. J. Cancer **35**:491, 1939.
13. Bell, E. T.: Am. J. Path. **19**:901, 1943.
14. (*a*) Geever, E. F.; Neubuerger, K. T., and Davis, C. L.: Am. J. Path. **19**:913, 1943. (*b*) Geever, E. F.; Carter, H. R.; Neubuerger, K. T., and Schmidt, E. A.: Radiology **44**:319, 1945. (*c*) Neubuerger.[4]

One hesitates to coin a new term to designate any condition in the nomenclature of which so much confusion already exists. However, "adenomatosis" is a proper designation for tumors which fulfil the three criteria enumerated, and "cancerous adenomatosis," for those which metastasize in addition. The descriptive phrase "epithelization of the alveolar walls" is also regarded as inappropriate, for many authorities deny that the adult pulmonary alveoli are lined by cells of epithelial origin. Therefore, "investment of alveoli" has been substituted, deliberately evading any speculation concerning the origin of the cells which line the alveoli in abnormal conditions such as pulmonary adenomatosis.

MATERIAL AND METHODS

The material on which this study was based has been derived from 9 cases collected in the files of the Army Institute of Pathology from military and civilian sources. Four additional cases were rejected because the data were incomplete or the postmortem observations controversial.

In every case in which the material was sufficient the following stains were used: hematoxylin and eosin, Masson's trichrome, Weigert's elastic and mucicarmine. In a few the Brown-Brenn and Giemsa stains were used. All sections were cut from formaldehyde-fixed tissues and paraffin blocks; however, in 1 instance additional celloidin (pyroxylin) sections were made for comparative purposes.

The Masson and elastic stains adequately demonstrated that the normal components were present in the alveolar walls. The Masson stain was superior to hematoxylin and eosin for the study of the structure of the investing cells. The mucicarmine stain revealed that the cells were secretory in several instances; but it is believed that when mucin was not seen, the tissue may have been washed free of it by long immersion in fixative. No bacteria were noted with the special stains. In our experience paraffin sections maintain the delicate cellular investment of the alveolar walls with no more distortion than is seen in sections made from celloidin-impregnated tissue.

CASES FROM THE ARMY INSTITUTE OF PATHOLOGY

CASE 1 (contributed by Arturo R. Casilli, M.D., Elizabeth, N. J.).—A white man aged 59 was admitted to a hospital with the history of sudden pain in the chest followed by increasingly severe dyspnea. Roentgenograms revealed discrete, nodular infiltration throughout both lung fields, and the clinical impression was pulmonary tuberculosis. There was progressive loss of weight and weakness, and the patient died approximately eight months after the onset of symptoms.

At autopsy, the right lung was firmly adherent throughout its entire posterolateral aspect. There were no adhesions of the left lung. The pleural cavity contained about 100 cc. of clear yellow fluid. The cut surface was studded with nodules, many of which were confluent (fig. 1). No mestastases were noted.

Microscopic Observations.—The alveolar spaces were lined by cuboidal to columnar cells (figs. 2 and 3). The nuclei were situated close to the cell bases; the nuclear membrane was sharply defined and the nucleolus was small, round, eccentrically placed and hyperchromatic. The chromatin was finely distributed, and no mitotic figures were present. The cytoplasm was eosinophilic and

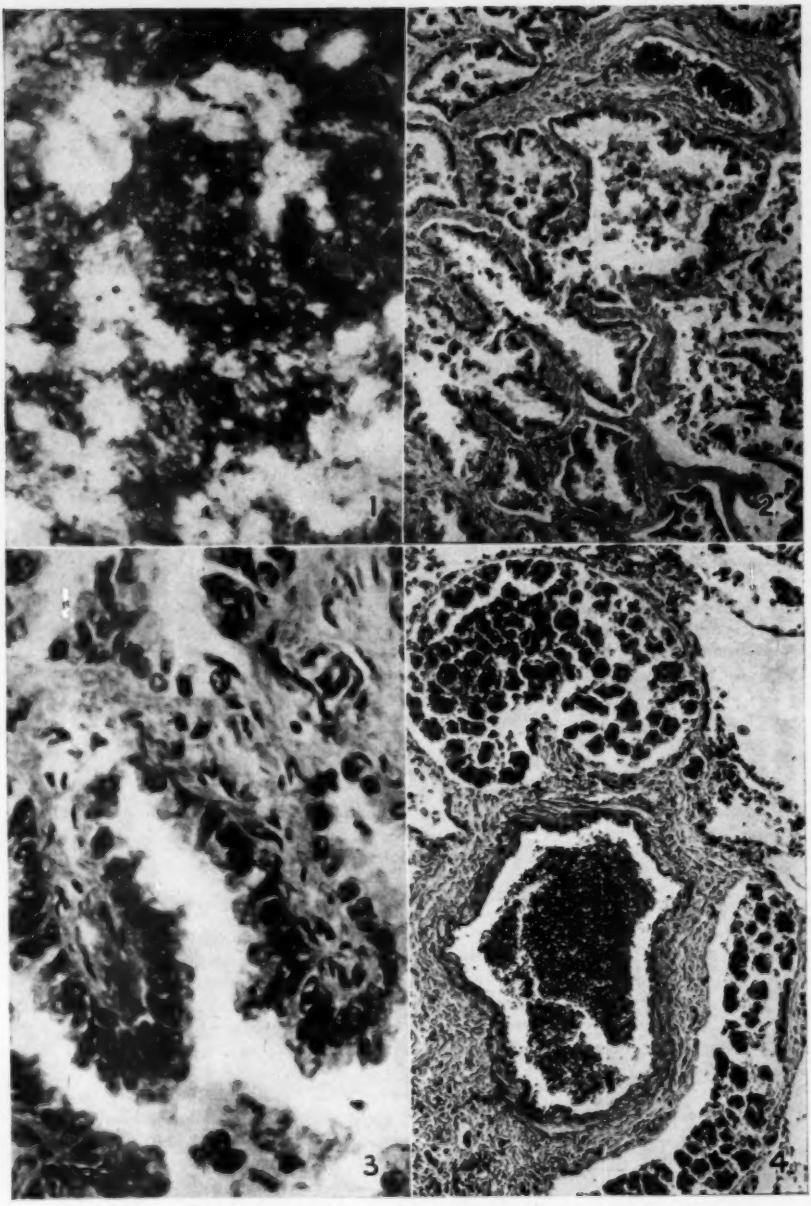

Fig. 1.—Cut surface of the right lung. There are numerous discrete and coalescing nodules. Army Institute of Pathology negative 99355.

Fig. 2.—Alveolar walls are slightly thickened and lined by cuboidal and columnar cells. There are occasional papillae and foci of desquamated lining cells. × 145. Army Institute of Pathology negative 85106.

Fig. 3.—Detail of investing cells. Note formation of papillae. × 658. Army Institute of Pathology negative 85099.

Fig. 4.—Papillary proliferation of atypical cells in perivascular lymphatic channels. × 130. Army Institute of Pathology negative 85107.

prominently vacuolated. Numerous delicate papillae partially filled the alveolar spaces. There were also many rounded individual cells, which were phagocytic, and rare polymorphonuclear leukocytes in the alveoli. The alveolar walls were intact, and there was no evidence of necrosis.

The medium-sized branches of the pulmonary artery demonstrated reduplication of the internal elastic lamina. The peribronchial and perivascular lymphatic channels were lined by cells similar to those occupying the alveolar walls (fig. 4).

CASE 2.—A white man, aged 40, when first seen complained of pain in the chest and cough. His temperature was 102 F., and acute bronchitis was diagnosed. Four months later he was seen again. Coughing had increased, and he expectorated small quantities of thick white phlegm. Roentgenograms of the chest revealed consolidation of the lower lobe of the right lung. Sputum became copious and was not blood streaked. Seven months after the initial symptoms the patient was producing from 1½ to 2 cups of mucoid sputum daily, in which no blood was noted. He became progressively weaker, with marked dyspnea on exertion, and died approximately eight months after he first was admitted to the hospital.

At autopsy the lungs were voluminous, filling the pleural spaces. There were bilateral adhesions with no loculated fluid. The right lung weighed 1,340 Gm. The lower lobe was firm, and the cut surface was studded with soft white nodules varying in size from bare visibility to a diameter of 2.0 cm. The left lung weighed 850 Gm. and was crepitant throughout, and there was no gross evidence of tumor. No necrosis or evidence of metastasis was noted.

Microscopic Observations.—The alveolar spaces were lined by a single row of nonciliated tall cylindric cells (figs. 5 and 6). The cells formed a striking pattern with their orderly array, lack of hyperchromatism, mitotic figures and lack of pleomorphism. The numerous papillary processes had delicate central stalks containing tiny capillary vessels. A bright red nucleolus, slightly eccentrically placed, was visible with Masson's stain. There were many mucin-filled vacuoles in the cytoplasm. The alveolar walls had been ruptured by massive intra-alveolar cellular proliferation. The normal mural components could be identified with special stains. No metastases were noted.

CASE 3.—A 31 year old white man had undergone two episodes of "flu," one in 1941, another in 1942, spending a week in the hospital on each occasion. Two months after the second episode he noted dyspnea and cough on slight exertion. The roentgenologist's report described scattered areas of exudative infiltration throughout the entire right lung and parts of the left, and the diagnosis was "far advanced tuberculosis." Repeated examination of sputum failed to reveal acid-fast organisms or fungi. The patient's course continued downhill, with progressively severe dyspnea, orthopnea, weakness and cough. Among the diagnoses considered were pulmonary tuberculosis, Boeck's sarcoid and histoplasmosis. The patient died three months after the onset of symptoms.

At autopsy, the left lung weighed 700 Gm. It cut with what was described as a "gritty resistance," disclosing numerous discrete and confluent, poorly circumscribed red-gray nodules. The right lung resembled the left. No metastases were noted.

Microscopic Observations.—Although extensive areas of the pulmonary parenchyma had been obliterated by fibrosis, there were many islands of surviving alveoli. The alveolar walls were lined with a single layer of tall cylindric cells, many of which were ciliated (fig. 7).

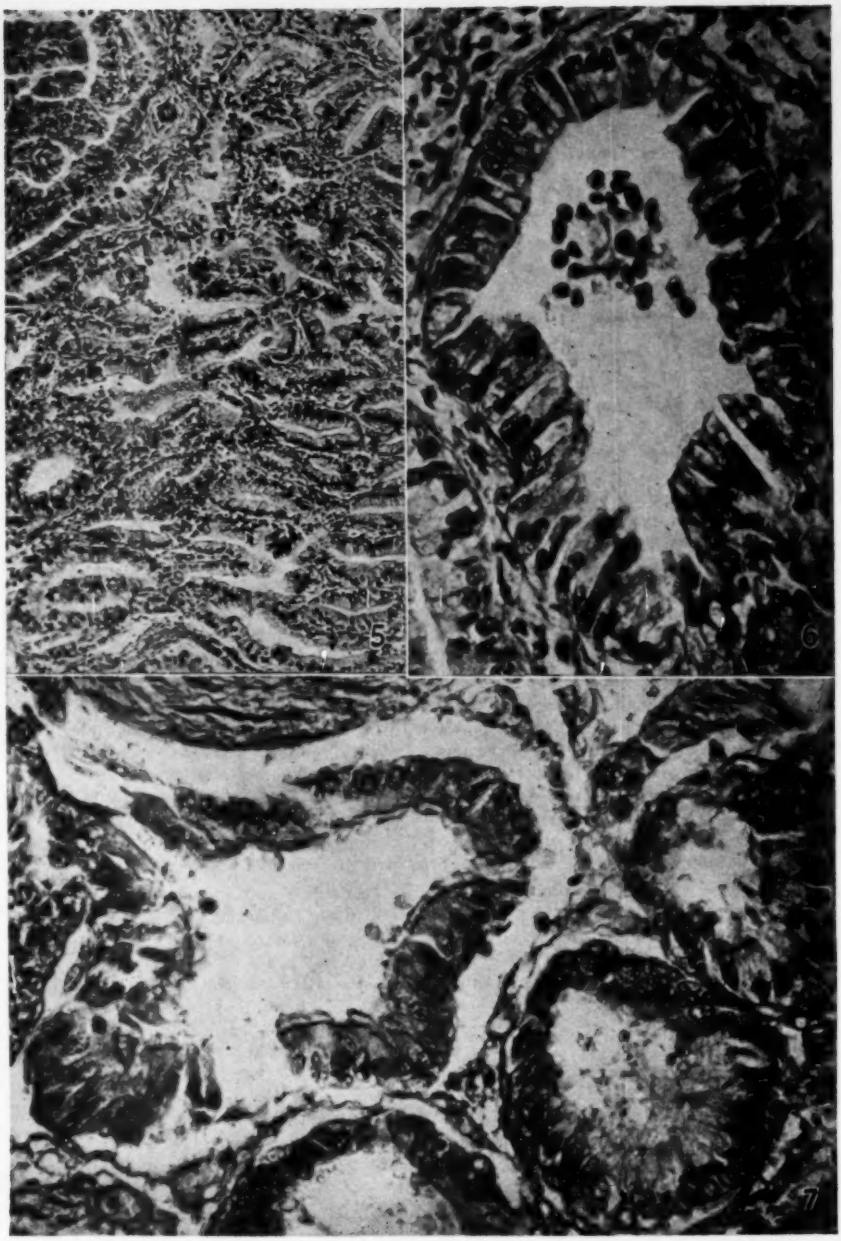

Fig. 5.—Alveolar walls are lined by tall cylindric cells. Papillary processes are frequent. × 100. Army Institute of Pathology negative 85110.

Fig. 6.—Detail of nonciliated columnar investing cells. Note intracytoplasmic vacuoles which contain mucin. × 500. Army Institute of Pathology negative 85100.

Fig. 7—Thickened alveolar walls lined by ciliated cylindric cells. × 300. Army Institute of Pathology negative 85098.

The cytoplasm contained secretory granules, and the alveolar spaces were filled with mucoid material in which there were many mononuclear cells and lymphocytes, moderate numbers of polymorphonuclear leukocytes and occasional multinucleated monster giant cells. There was marked peribronchiolar inflammatory cell infiltration. No evidence of metastasis was noted.

Case 4 (contributed by David M. Grayzel, M.D., Brooklyn).—The patient was a 43 year old white man. During a fluoroscopic examination he was found to have a "spot" on the right lung. He was treated twelve times with high voltage roentgen rays, presumably for carcinoma of the lung. He had had a cough for an undetermined time, which became severe ten days before he was admitted to the hospital. Roentgenograms of the chest revealed an opacity obscuring the right lower lung field. Pneumonectomy was performed on the right side. Six months later the patient died with massive pleural effusion. Permission for autopsy was not granted, but there was no clinical evidence of recurrence of tumor.

The right lung, removed at operation, weighed 1,240 Gm. The lower lobe was voluminous, grayish pink and white, and of a rubbery consistency. The cut surface was studded with closely approximated grayish yellow nodules (fig. 8). A description of the left lung is unavailable.

Microscopic Observations.—The alveoli were lined by nonciliated tall cylindric cells, and the spaces were almost solidly filled with papillary processes. The nuclei were situated at the bases of some cells and at the apexes of others. Those cells having the nuclei situated at the free ends were distinctly club shaped. There was abundant cytoplasm with various-sized vacuoles containing material which stained red with mucicarmine. In many alveoli a delicate wrinkled membrane lay in close approximation to the alveolar wall.

The alveolar walls were intact and acted as a supporting stroma for the lining cells. The intramural capillaries were engorged and contained many polymorphonuclear leukocytes. Several areas of lung parenchyma contained sheets of polymorphonuclear leukocytes associated with microabscesses.

The bronchioles were surrounded by alveoli lined with investing cells; however, the bronchiolar epithelium consisted of normal cells. Dr. Nathan Chandler Foot commented on these sections as follows: "Bronchogenic adenocarcinoma. Might well have originated in pulmonary adenomatosis."

Case 5.—A 66 year old white man complained of long-standing productive cough, pain in the chest and dyspnea on exertion. There had been hemoptysis for three months prior to hospitalization. A roentgenogram revealed atelectasis of a portion of the lower lobe of the right lung. Pleural fluid was obtained and an examination made for cancer cells and acid-fast organisms; none was found. A bronchoscopic examination revealed moderate secretion from the bronchus of the lower lobe of the right lung and no evidence of tumor. The patient was discharged from the hospital. At home he continued to be troubled with a cough productive of a moderate amount of mucoid material. His condition did not improve, and he suddenly died after approximately fifteen months' illness.

At autopsy there were a few fibrous adhesions and no effusion in the right pleural space. The left pleural space contained 300 cc. of clear straw-colored fluid and occasional bandlike adhesions were noted at the apex and the lower lobe of the left lung. The interlobar fissures were obliterated. The right lung weighed 1,287 Gm. The cut surface of the entire lung was gray and consolidated

except at the periphery, where there were numerous small nodules. The left lung weighed 2,015 Gm. The cut surface was similar to that of the right lung.

The bronchi of both lungs appeared to be normal, and no metastases were noted.

Microscopic Observations.—There was an increase in the amount of fibrous tissue throughout the bronchial wall, marked peribronchial fibrosis and moderate hyperplasia of the mucous glands, but no evidence of bronchogenic tumor. The alveoli surrounding the bronchus were lined by nonciliated cuboidal and columnar cells (fig. 9). The investing cells projected irregularly into the alveolar spaces with bulbous or whiplike processes, often containing several nuclei. There were many mucus-filled cytoplasmic vacuoles. The alveolar walls were delicate and formed a latticework for the neoplastic cells. There were extensive areas of fibrosis, and special stains revealed marked obliterating endarteritis of the medium-sized pulmonary arteries. No metastases were noted.

Case 6.—A white man 35 years of age was observed, by routine roentgenogram of the chest, to have evidence of pulmonary disease. He had had no premonitory signs or symptoms and was completely unaware of the existence of pulmonary disease. Lobectomy of the left lower lobe was performed, and no extension to the adjacent lung or to the regional lymph nodes was noted. The patient's immediate postoperative course was uneventful.

In June 1947, roentgenograms of the chest revealed lesions in the upper and middle lobes of the right lung. A specimen was taken from the upper lobe for biopsy, and Dr. Sidney Farber immediately inoculated laboratory animals, including sheep and guinea pigs, with some of the tissue and planted some in culture mediums. No growth was obtained on culture, and no evidence of disease was noted in the inoculated animals.

Biopsy showed that the material examined consisted of the surface of the lower lobe, which was mottled gray and black and showed no adhesions. On cut surface several nodules measuring up to 2.5 cm. were encountered. These were present in the subpleural region, and one was found lateral to the main stem bronchus but did not communicate with it. The surface of the nodules was smooth, glistening, gelatinous, and divided into coarse trabeculations. Dissection of the bronchi revealed no abnormalities, and several hilar lymph nodes appeared to be free of metastases.

Microscopic Observations.—There was a remarkable investment of the alveolar walls by large, tall columnar cells, which were actively secreting a mucoid material. The alveolar spaces contained this mucoid substance and many desquamated lining cells. In several places the production of mucus was so copious that the alveolar walls had ruptured, allowing pools of mucus to form (fig. 10). The pulmonary stroma was intact in all other areas. No metastases were noted in the sections of hilar lymph nodes or peribronchial lymphatic vessels.

Case 7 (contributed by Theodore L. Bliss,[15] M.D., Akron, Ohio).—A 47 year old white man first noticed an unproductive cough. He was a chemist and stated that he had long been exposed to sulfur dioxide and other chemical pulmonary irritants. Within four months the cough became productive. During the fifth month of his illness he became febrile and raised copious amounts of thin fluid. Roentgenograms of the chest revealed increased density in the region of the middle lobe of the right lung. Laboratory examinations did not reveal tubercle bacilli or fungi.

15. Bliss, T. L.: J. Thoracic Surg. **6**:660, 1936.

On bronchoscopic examination, profuse secretion was seen to well up from the bronchus of the lower lobe of the right lung, which had become narrowed. The quantity of bronchial secretion in a twenty-four hour period was prodigious and was estimated to vary between 1,200 and 1,800 cc. The secretion was watery, never contained blood or pus and had no odor and no taste.

The patient died approximately thirteen months after the onset of symptoms. At autopsy, the right pleural cavity was obliterated by dense fibrous adhesions, and there were a few easily separated adhesions in the left pleural cavity. On section, the entire right lung was seen to be replaced by grayish white tumor. Approximately one third of the left lung, in the vicinity of the hilus, was replaced by similar neoplastic tissue.

Microscopic Observations.—The alveolar walls were lined by nonciliated columnar cells, which appeared to be arranged in a pseudostratified fashion. The cytoplasm was strongly eosinophilic, and the nuclei were round to oval and were usually situated near the bases of the cells; however, where the lining cells were irregularly grouped, nuclei were situated near the tips of the cells. The alveolar walls were of normal width and furnished a lattice on which the proliferating cells formed papillary processes. The pulmonary septums were more prominent than normal and contained occasional lymphocytes, plasmacytes and polymorphonuclear leukocytes. No metastases were present in the peribronchial or pleural lymphatic channels.

Case 8.—A 62 year old woman complained of having a feeling of irritation in the posterior part of the pharynx, incapacitating dyspnea on exertion and persistent cough with production of small amounts of clear mucoid material, without hemoptysis.

Bronchoscopic examination revealed an anterior-posterior narrowing of the dorsal branch bronchus of the lower lobe of the right lung, and no endobronchial tumor was seen. Examination of pleural fluid obtained from the right cavity of the chest was reported as "carcinoma cells morphologically suggesting metastatic epithelial origin."

Roentgenograms revealed uniform density of the lower half of the right lung and a fine type of miliary infiltration throughout the upper portion of the right and the entire left lung.

Respiratory distress was extreme and constant. Weakness, cyanosis and dyspnea increased in severity, and the patient died approximately five months after the onset of symptoms.

At autopsy, the pertinent findings were limited to the lungs; no metastases were noted in the regional lymph nodes. The gross description of these lungs is unavailable.

Microscopic Observations.—The pleura was greatly thickened and fibrous, and scattered through it were small oval spaces resembling lymphatic channels lined by a single layer of large atypical cells. The alveolar walls were also lined by a layer of cells identical to those seen in the pleura. The alveolar walls were slightly wider than normal; however, special stains failed to demonstrate any fibrosis, and the elastic and reticular elements were normal. The neoplastic cells were present in the perineural (fig. 11), perivascular and peribronchial lymphatic channels. In most places these cells lined the lymphatic walls in a manner identical to that seen in the alveoli, and the lumens contained occasional desquamated cells. The structure of the peribronchial lymph nodes was partially obliterated by these cells. No mitotic figures were noted in the parenchymal or the lymphatic neoplastic cells.

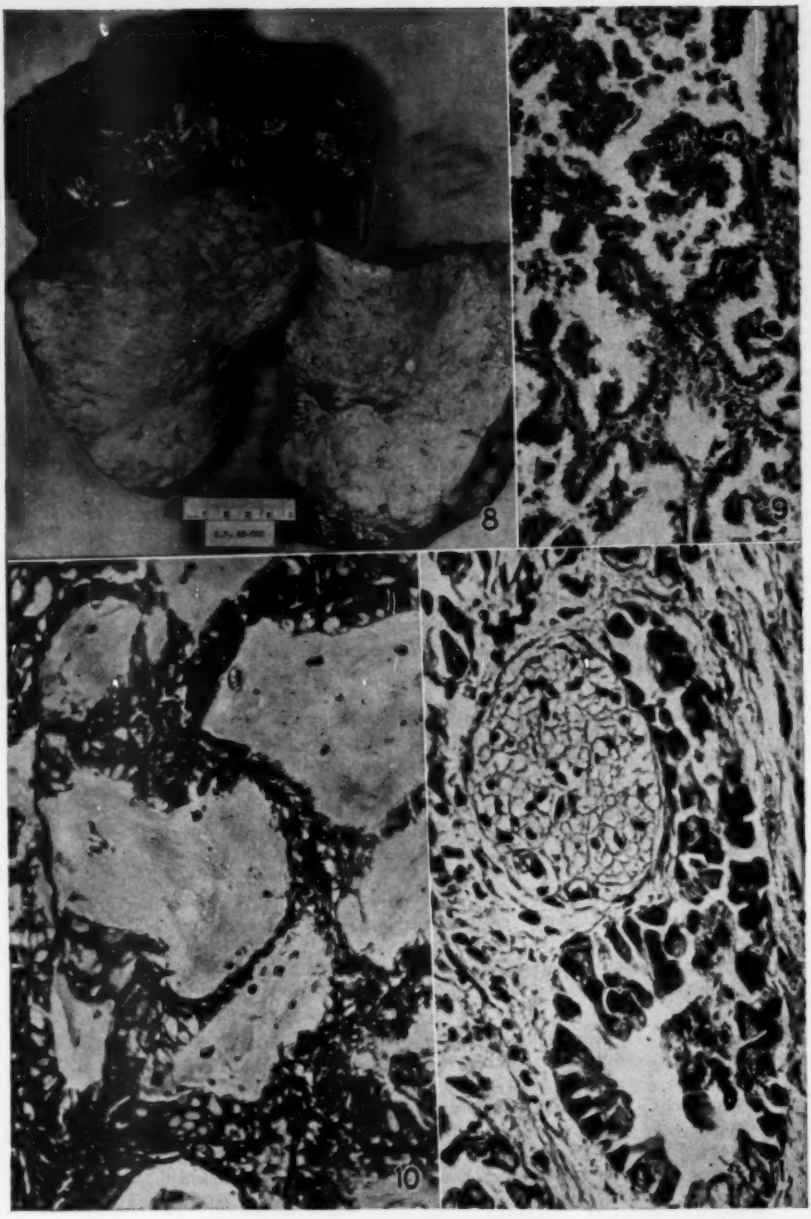

Fig. 8.—Diffuse involvement of the right lower lobe of the right lung. Note the smooth pleural surface and suggestion of peripheral nodularity on cut section. Army Institute of Pathology negative 98159.

Fig. 9.—Low power view showing investing alveolar cells. The alveolar walls are thickened. Note the remarkable similarity between this picture and that seen in jagziekte (fig. 14). Army Institute of Pathology negative 99404.

Fig. 10.—Pools of mucus in alveolar spaces. The investing cells have large intracytoplasmic vacuoles. × 390. Army Institute of Pathology negative 99399.

Fig. 11.—Perineural lymphatic vessels lined by cells identical with those investing the alveoli. × 450. Army Institute of Pathology negative 100365.

CASE 9.—A 42 year old white man was admitted to the hospital, complaining of shortness of breath and pain in the right side of the chest. It was believed that he had a tumor of the right lung, and bronchial biopsy was said to show tumor which probably arose in the gastrointestinal tract. Roentgenograms of the gastrointestinal tract failed to demonstrate any abnormalities. An exploratory thoracotomy revealed collapse of the lower lobe of the right lung and presence of nodules throughout the right leaf of the diaphragm.

The course of the disease was marked by effusion, requiring repeated aspirations of the right pleural cavity, and developing empyema. Cough was moderate, and 2 to 3 ounces (60 to 90 cc.) of sputum was produced daily. After a prolonged downhill course, the patient's condition deteriorated rapidly, and he died approximately fifteen months after the onset of symptoms.

At autopsy, the right pleural cavity contained a large quantity of semifluid greenish black material, and the left pleural cavity contained 3,300 cc. of thin amber-colored fluid. The cut surfaces of the lower lobes were gray, and dissection of the bronchial tree failed to demonstrate either endobronchial tumor or ulceration. The hilar and mediastinal lymph nodes were grossly invaded by tumor.

Tumor nodules were present over the entire surface of the right leaf of the diaphragm. The liver contained numerous gray nodules, which did not elevate Glisson's capsule and were not umbilicated. The right adrenal gland contained an 8 mm. tumor in the medulla. There were also metastases in the periaortic lymph nodes, and several implants were present on the serosa of the gastrointestinal tract.

Microscopic Observations.—Near the pleural-parenchymal junction numerous lymphatic channels were lined by atypical cells. The alveolar walls had been diffusely invested by similar cells. These cells were large, with eosinophilic granular cytoplasm. The nuclei were regular in size, and no mitotic figures were seen. In some areas the cells were clumped, giving the appearance of pseudogiant cells. Desquamated cells were present in the alveolar spaces, and large cytoplasmic vacuoles were common. The alveolar walls were of normal width, and there were occasional papillary processes.

The peribronchial lymphatic channels were filled with proliferating tumor cells, and the regional lymph nodes had been almost completely replaced by tumor tissue. The microscopic picture of the local and distant metastases (figs. 12 and 13) resembled that of the pulmonary parenchyma, and mitotic figures were rare.

ANALYSIS OF CASES FROM ARMY INSTITUTE OF PATHOLOGY

A review of the data of the 9 cases from the Army Institute of Pathology shows that the ages of the patients varied from 31 to 66 years, with an average age of 47.2 years. The sex incidence was 8 males to 1 female. The preponderance of males can be explained by the selective nature of the material acquired by the Army Institute of Pathology. All the patients were white.

The duration of the disease was extremely difficult to evaluate, but in 8 cases in which there were symptoms the duration from the onset of the symptoms varied from four to fifteen months. In case 6 there were no symptoms and the lesion was discovered by routine roentgenologic examination (see table 1).

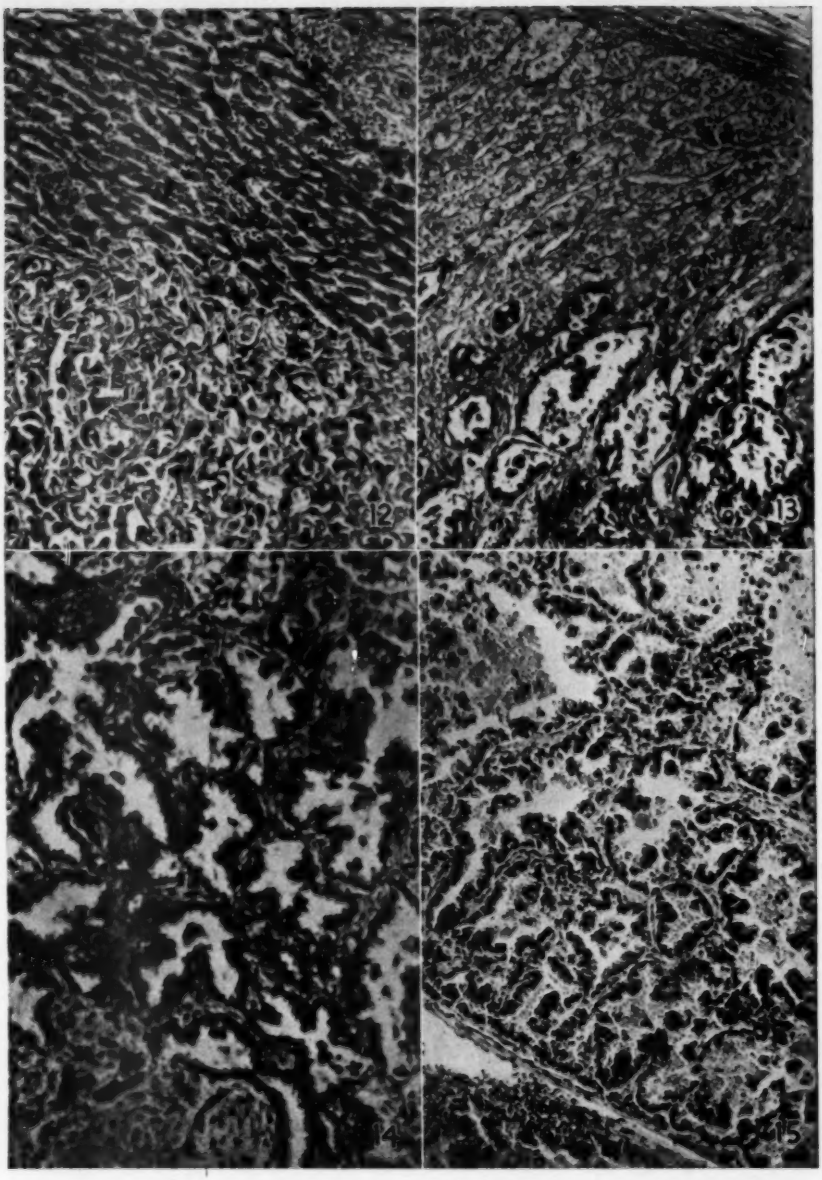

Fig. 12.—Metastasis in liver. The cells show considerable anaplasia; however, mitotic figures are rare. × 114. Army Institute of Pathology negative 100382.

Fig. 13.—Metastasis in adrenal gland. Note the similarity of pattern between the metastasis and the growth seen in the lung. × 114. Army Institute of Pathology negative 100381.

Fig. 14.—Jagziekte in sheep. Note the similarity between this and human pulmonary adenomatosis (fig. 9). × 160. Army Institute of Pathology negative 99551.

Fig. 15.—Caprine pulmonary adenomatosis. × 115. Army Institute of Pathology negative 100379.

(Contributed by C. L. Davis, DVM.)

The history of pulmonary irritants was established in case 7 only; the patient had been exposed to irritating chemical fumes in a fertilizer plant for many years. One patient (case 2) had been a cab and truck driver and may have been exposed to exhaust fumes. In most of the remaining cases a history of repeated bouts of "flu" or colds was given. Only in case 5 was there a history of heavy smoking.

Signs and symptoms in order of frequency were: productive cough, fever, dyspnea, weakness, loss of weight, thoracic pain, fatigue, cyanosis, night sweats, pleural effusion and clubbing of the fingers.

The usual clinical impressions in order of frequency were: tuberculosis, bronchogenic carcinoma, bronchopneumonia, bronchiectasis, Boeck's sarcoid, histoplasmosis and coccidioidomycosis.

TABLE 1.—*Nine Cases of Pulmonary Adenomatosis Discovered by Screening Material at the Army Institute of Pathology*

Case	Army Institute Pathology Accession	Sex	Age	Race	Location	Source of Material	Metastases	Duration, Mo.
1	58048	M	59	W	Bilateral	Autopsy	Lymphatic channels	8
2	96875	M	40	W	Right lung	Autopsy	None	4
3	107356	M	31	W	Bilateral	Autopsy	None	4
4	132190	M	43	W	Lower lobe, right lung	Pneumonectomy	Rarefaction in left tibia and questionable metastases in mesenteric nodes	12
5	147459	M	66	W	Bilateral, esp. lower lobe, right lung	Autopsy	None	15
6	165404	M	35	W	Lower lobe, left lung	Lobectomy	None	2
7	170824	M	47	W	Bilateral, mostly lower lobe, right lung	Autopsy	None	18
8	174836	F	62	W	.....	Autopsy	Bronchial lymph nodes	5
9	105690	M	42	W	Bilateral	Autopsy	Metastases in diaphragm, liver, adrenal gland, periaortic lymph nodes and peritoneum	15

The pathologic material was obtained from seven autopsies, one pneumonectomy and one lobectomy. The patient on whom pneumonectomy was done died six months after operation, but permission for autopsy was not granted. The patient on whom lobectomy was done has survived to the date of this writing.

The distribution of the tumor, whether nodular or diffuse, was not always apparent from the descriptions of the gross specimens, and the impression gained was that in most cases the process was actually a coalescence of nodules. There was no necrosis of the tumor. Metastases were noted grossly in only 1 case.

Histologically, these cases fulfilled all the criteria for pulmonary adenomatosis. Metastases were limited to the parenchymal lymphatic

channels in 1 lung (case 1) and to parenchymal lymphatic channels and hilar lymph nodes in 1 (case 8); there was widespread metastasis only in case 9. In the patient on whom pneumonectomy was done, there was a suspicion that metastases were present, but in the absence of autopsy confirmation was not possible.

ANALYSIS OF COLLECTED CASES

The data of cases described in the available literature were compiled by Neubuerger and Geever[1] in 1942, and among the tumors were 25 which these authors regarded as unmistakable "alveolar cell tumors." The ages of the patients varied from 20 to 89 years, and the sex ratio was given as approximately 1:1. A review of a majority of these cases has revealed that they fulfil the criteria given for pulmonary adenomatosis.

Twenty-six case reports have appeared in the literature since 1941, and I have added one[16] which had been published prior to 1941 but which Neubuerger and Geever did not include in their review. In table 2 are recorded 12 of these cases of pulmonary adenomatosis in which no metastases were found, and in table 3, 15 cases in which metastases had occurred. The 27 cases in tables 2 and 3 adequately fulfil all criteria previously enumerated. Table 4 lists 3 cases in which the condition described may be considered doubtful pulmonary adenomatosis and which are therefore not included in the following analysis.

Analysis of the 27 acceptable cases reveals that the ages of the patients varied from 17 to 79 years. Ten were between 50 and 59; 5, between 40 and 49, and 5, between 60 and 69. Thus 20 of the tumors (74.1 per cent) occurred in the 40 to 70 year age group. Twelve patients (44.4 per cent) were men and 15 (55.6 per cent) were women. Evidence of invasion or metastasis was present in 15 (table 3), or 55.6 per cent.

A comparison was made of 100 patients with bronchogenic carcinoma of the lung whose records are in the possession of the Army Institute of Pathology and the 27 patients with pulmonary adenomatosis reported in the literature and reviewed here, which revealed that the latter made up a considerably older group. The difference can be explained, however, by the fact that the two sets of cases are samples drawn from two populations known to be different, Army and civilian, respectively. The relative predominance of this condition in the female sex is remarkable when one considers that in series of cases of bronchogenic carcinoma the ratio of men to women varies from 3:1[17] to 10:1.[18] Therefore the

16. Sayago, G.: Prensa méd. argent. **19**:545, 1932.
17. Arkin, A., and Wagner, D. H.: J. A. M. A. **106**:587, 1936.
18. Grove, J. S., and Kramer, S. E.: Am. J. M. Sc. **171**:250, 1926.

TABLE 2.—*Cases of Pulmonary Adenomatosis Without Metastases Collected from the Literature*

Author	Age	Sex	Thoracic Findings	Authors' Interpretation
Sayago,[16] 1932	33	M	No pleural adhesions. Both lungs congested, gray hepatization throughout lower lung fields; miliary nodules, many confluent, scattered throughout lower lobes Microscopic: Alveolar walls invested with mantle of cuboidal and cylindric cells; papillae frequent; no mucus demonstrated	"Belongs to group of tumors arising from alveoli"
Sims,[24a] 1943	42	M	In each pleural cavity, about 100 cc. of serosanguinous fluid. Adhesions present. Lungs voluminous; multiple nodules bilaterally, most extensive in right lung Microscopic: Alveoli lined by single layer of high cuboidal cells; alveolar walls normal	". . . the alveolar exudate and the epithelial hyperplasia . . . are quite like those in the jaggiekte sections. . . ."
Bell,[13] 1943	63	M	Diffuse consolidation of lobes of both lungs Microscopic: Alveoli lined by cuboidal and columnar cells; alveolar walls and pulmonary septums essentially normal	"There is convincing evidence that the epithelium forms locally and does not grow from bronchi"
Taft and Nickerson,[4c4] 1944	62	M	Numerous fibrinous adhesions bilaterally. Left lung 900 Gm., firm, consolidated; cut surface of upper lobe grayish white. Right lung 850 Gm., almost completely consolidated Microscopic: Alveolar walls covered by columnar cells and occasional papillae	"While a definite conclusion cannot be made, it seems probable that the abnormal cells arise from the alveolar lining"
	79	F	Right lung 700 Gm.; on section lower and middle lobes were gray centrally and dark red peripherally. Left lung 420 Gm.; on section lower lobe gray, gelatinous Microscopic: Alveoli lined by columnar cells; surrounding alveoli filled with mucoid secretion and desquamated cells	
Wood and Pierson,[20] 1945	57	F	Right lung on section had multiple small grayish miliary nodules. Left lung similar Microscopic: Alveoli lined by columnar cells forming papillae; stroma intact	. . . "columnar epithelial cells appear to arise de novo from the alveolar walls"
Ikeda,[4a] 1945	50	F	In each pleural cavity from 2,000 to 3,000 cc. of clear fluid. Right lung 500 Gm.; left, 750 Gm.; on section parenchyma was studded with whitish tumors Microscopic: Papillary growth arising from the alveolar lining	. . . "the tumor can originate from the lining cells of the alveoli and is epithelial in character"
Geever, Carter, Neuburger and Schmidt,[14b] 1946	17	F	Left lung and middle and lower lobes of right lung occupied by grayish tumor nodules varying from pinhead size to diameter of 1.0 cm. Microscopic: Alveolar walls lined by columnar cells with rare mitotic figures	"Although the majority of alveolar-cell tumors have been definitely malignant, some were borderline or histologically benign"
Paul and Ritchie,[21] 1946	68	F	Lower lobe of left lung studded with 2 mm. nodules. Bronchiectatic cavities present bilaterally	"It appears . . . that the abnormal epithelium of adenomatosis is derived, in some cases at least, from bronchial epithelium"
Deserman and Neuhof [10]	44	F	Lobectomy, lower lobe, left lung; on section lung tissue completely replaced by grayish tumor; bronchi pushed toward pleural surface; no endobronchial tumor seen Microscopic: Basement membrane of tumor consisted of alveolar walls; cells cylindric, mucus containing	
Alexander, C. M., and Foo Chu: Arch. Path. 43 : 92, 1947	56	F	Adhesions in right pleural space and none in left. Lungs together weighed 2,100 Gm. Lower lobe of right lung voluminous, yellowish-pink, with gelatinous cut surface. Solitary nodule in left lung Microscopic: Tall columnar cells; alveoli, noncellated, no mitotic figures, droplets of mucin in cytoplasm. Nearly every alveolus contained fibrinopurulent exudate	"We believe that in our case the tumor was most likely of multicentric origin since there was no demonstrable invasion of lymphatic vessels"
Simon, M. A.: Am. J. Path. 23 : 413, 1947	70	F	Right thoracic cavity partially obliterated by fibrous adhesions; less extensive adhesions on left. Cut surfaces gray, consolidated. Friedländer's bacilli cultured Microscopic: Majority of alveoli filled with polymorphonuclear leukocytes; walls lined by single or pseudostratified layers of tall columnar epithelial cells	"Pulmonary adenomatosis and alveolar cell tumors may be regarded as unusual forms of pulmonary carcinoma presumably arising from alveolar lining cells"

TABLE 3.—*Cases of Cancerous Pulmonary Adenomatosis Collected from the Literature*

Author	Age, Sex	Thoracic Findings	Metastases	Authors' Interpretation
Smith and Gault,[46b] 1942	48 M	Right visceral pleura studded with grayish nodules. Complete consolidation of right lung resembling hepatization Microscopic: Alveolar walls show moderate fibrosis and capillary dilatation; lining cells large and polyhedral	Right visceral pleura, pulmonary lymphatic channels, peribronchial and peritracheal lymph nodes	"Indication of possibility that cells are of actua alveolar lining cell origi with tendency toward adenocarcinoma"
Dacie and Hoyle,[11] 1942	54 M	Pleural cavities partially obliterated bilaterally. Both lower lobes, right middle lobe, left upper lobe and lingula diffusely affected by changes suggesting chronic confluent bronchopneumonia Microscopic: Epithelial proliferation within existing alveolar structure; alveolar walls intact; papillae numerous; in some areas, mucin-secreting cells	Invasion of pleura	. . . "they (tumors) appea to arise from alveoli o the lung rather than b extension from the bron chioles or bronchi"
Wood, E. H., Jr.: Radiology 40:193, 1943	62 F	Lungs appeared to be in state of diffuse gray hepatization, composed of lobular pinkish tan tumor nodules Microscopic: Alveoli lined by simple or pseudostratified columnar tumor cells; few mitotic figures; numerous papillae; local invasion and destruction of alveolar septums	Local invasion and destruction of alveoli	Inclined to feel there is som relation between lipoid pneumonia and carcinom arising from epithelial lin ing cells of lung
Herbut,[45a,b] 1944....	50 F	Bilateral adhesions. Entire right lung consolidated and gray; cut surface homogeneous gray, with nodules at periphery; area of early necrosis in upper lobe. Left lung studded with circumscribed and confluent nodules Microscopic: Alveoli lined with one or more layers of cuboidal or columnar cells and septums thickened	Peribronchial and perivascular lymphatic channels, pericardium, mediastinal lymph nodes, liver, adrenal glands and vertebral marrow	"It is believed that the parent cell in all cases o primary carcinoma of th lung is the basal cell o the bronchial or bronchio lar mucosa. The distribu tion of the subsequent tumor is dependent upon the further differentiatio of the cells. . . . If the are cuboidal or columnar they will regularly line th septa producing the well known alveolar arrange ment . . ."
	48 F	Adhesions in upper right pleural cavity. Entire right lung diffusely consolidated with grayish white tumor tissue. Left lung studded with pinhead size grayish nodules Microscopic: Alveoli regularly lined by tall cuboidal cells	Mediastinal lymph nodes, pericardium and liver	
	52 F	Adhesions in left pleural space. Entire left lung infiltrated with discrete and coalescing gray nodules. Right lung essentially similar Microscopic: Tall columnar cells lined alveoli; considerable sloughing of cells in alveolar spaces	Tumor cells present in lymphatic vessels	
Ikeda,[46a] 1945.....	52 F	On section right lung consolidated with grayish white tissue from region of hilus. Left lung studded with nodules Microscopic: Papillary growth arising from alveolar lining; cells tall columnar	Both lungs regional lymph nodes, spleen, liver, adrenal glands, spine and pelvis	. . . "The path leading t the fully developed alveo lar cell carcinoma of th lung may be assumed t begin as local hyperplasia
	60 F	(No description given)	Regional lymph nodes and pleura	often in multiple center later developing into be nign adenomatosis and finally becoming carci nomatous"
Geever, Carter, Neuhuerger and Schmidt,[24b] 1945	49 M	Both lungs studded with firm gray nodules, occasionally coalescent Microscopic: Nodules composed of anaplastic cells lining alveolar walls and forming papillae	Liver, adrenal glands, hilar lymph nodes	"Although the majority o alveolar-cell tumors hav been definitely malignant some were borderline o histologically benign"
	53 F	Both lungs studded with gray nodules which occasionally encroached on pleura Microscopic: Alveolar walls thickened and lined by cuboidal cells	Brain, pulmonary lymphatic vessels	
	76 M	Left lung occupied by grayish yellow and white tumor nodules Microscopic: Alveoli lined by highly anaplastic cells with many mitotic figures; scattered areas of necrosis	Regional and peripancreatic lymph nodes; pancreas, adrenal glands, and kidneys; masses of tumor cells in lymphatic channels	
	58 M	Left pleura studded with nodules. On section upper lobe of left lung solid, pale gray; lower lobe contained a few nodules. No tumor in right lung Microscopic: Alveolar walls lined by cuboidal or cylindric cells with occasional mitotic figures and giant cells	Implants on pleura and pericardium; lymphatic channels contained tumor cells	

TABLE 3.—*Cases of Cancerous Pulmonary Adenomatosis Collected from the Literature—Continued*

Author	Age, Sex	Thoracic Findings	Metastases	Authors' Interpretation
Fishman, A. P.; Epstein, B. S., and Grayzel, D. M.: Am. Heart J. **30**: 809, 1945	20 F	Right lung firm and rubbery throughout; nodules in left lung; on section it resembled pneumonic consolidation; no necrosis. Left lung revealed a few circumscribed nodules Microscopic: Alveoli filled with tumor cells; stroma of tumor made up of alveolar walls; rare mitotic figures; marked endarteritis	Nodules in left lung	
Wenger,[46c] 1945.....	60 F	Adhesions present bilaterally. On section right lung firm, grayish yellow, with appearance of hepatization. Left lung studded with nodules, confluent in upper lobe Microscopic: Alveoli lined by epithelial cells, with occasional formation of papillae	Liver	Tumor appeared to be multicentric and most likely originated in cells lining pulmonary alveoli
Simon, M. A.: Am. J. Clin. Path. **17**: 783, 1947	39 M	Right pleural cavity completely obliterated by dense adhesions. Cut surface of right lung composed of pinkish gray tumor tissue. Left lung also involved Microscopic: Alveoli lined by single and sometimes pseudostratified layers of tall columnar cells; papillae common; mitotic figures rare. Metastases were identical with tumor in lung	Brain	"Although controversial, the evidence suggests that this tumor is multicentric in origin and is probably derived from lining epithelial cells"

reversed sex incidence of 1 : 1.25 in this series of cases of adenomatosis resembles that of bronchial adenoma more closely than that of bronchogenic carcinoma.[19]

It appears, moreover, that the statement that the majority of alveolar cell tumors are definitely cancers [14b] is borne out by these data. Neuhuerger and Geever [1] estimated that 25 per cent metastasize to the regional lymph nodes and another 23 per cent to distant sites. It is possible that thorough histologic examination of the peribronchial and hilar nodes would reveal that the suggested incidence of metastases is too conservative.

THE CHARACTERISTIC LESION

The following gross and microscopic descriptions are a composite of details drawn from the literature and from the material studied at the Army Institute of Pathology.

Gross Description.—When death has resulted from pulmonary adenomatosis, the pleural space is usually partially or completely obliterated by fibrous adhesions. The pleural cavity sometimes contains fluid, which is usually clear and may total as much as 3,300 cc. (case 9). The lungs may vary considerably in weight, recorded weights ranging from 356 Gm.[20] to 2,750 Gm.[5] The lungs are voluminous and tend to maintain their contours when the chest is opened. The visceral and parietal pleurae may be studded with gray to grayish pink nodules.

19. Clerf, L. H., and Bucher, C. J.: Ann. Otol., Rhin. & Laryng. **51**:836, 1942. Holly, S. W.: Mil. Surgeon **99**:528, 1946.

20. Wood, D. A., and Pierson, P. H.: Am. Rev. Tuberc. **51**:205, 1945.

It has become a common practice to describe the gross distribution of these tumors as nodular (miliary), diffuse or a combination of these two forms. Distribution may vary from involvement of a single lobe of one lung to all lobes of both lungs.

In the nodular variety, the cut surface is studded with tumor foci, ranging from minute to coalescing nodules, which may occupy an entire lobe. Peripherally the larger areas are irregular, and cut surfaces are yellowish white to grayish pink. The consistency is soft and often distinctly mucoid.

The diffuse form is characterized by homogeneous involvement of extensive areas of the parenchyma, but a suggestion of nodulation often is noted at the periphery of the neoplasm. Areas of necrosis of the tumor are exceedingly rare, but changes due to superimposed infection may simulate necrosis of tissue.

TABLE 4.—*Doubtful Cases of Pulmonary Adenomatosis Collected from the Literature*

Author	Age, Sex	Thoracic Findings	Metastases	Author's Interpretation
Sanda, E.: Casop. lék., cesk. 80: 237, 1941	65 F	Pulmonary tissue firm and on section resembled pneumonic hepatization Microscopic: "Cancerous pneumonia"	.....	Alveolar carcinoma
	43 M	(No description given)	Mediastinum	Alveolar carcinoma
Ikeda,[46a] 1945.....	56 M	Numerous flat nodules on pleura of right lung. On section, surface of right lung studded with grayish white nodules. Left lung essentially the same	Secondary bronchus, (obstructed by growth), brain and meninges	Alveolar cell carcinoma of lung

Intervening lung tissue is often hyperemic and shows evidence of inflammation. On compressing the lung, mucinous material exudes, sometimes associated with pus, for multiple abscesses and bronchiectasis occasionally complicate the picture. The gross appearance may be easily confused with that of the chronic granulomas of the lung which occur in tuberculosis, coccidioidomycosis and histoplasmosis, or with that of secondary neoplasms, especially those arising in the gastrointestinal or the genitourinary tract, or with that of such conditions as Boeck's sarcoid, leukemic infiltration or parasitism. The gross specimens of lungs available for study in this group often suggested the gray hepatization of lobar pneumonia. On the other hand, on gross examination a case of extensive areas of unresolved pneumonia was misinterpreted as one of adenomatosis.

Microscopic Description.—The histologic picture is essentially the same in both the nodular and the diffuse form. There are variations in the pattern, ranging from simple investment of alveoli to complicated

arrangements resulting from extensive intra-alveolar proliferation with rupture of alveolar walls and coalescence of spaces.

In the simplest form the alveolar walls are lined by a single layer of cuboidal or cylindric cells, which are usually nonciliated.[21] The cells are remarkable for their monotonous regularity of structure and arrangement. The cytoplasm is faintly eosinophilic, and special stains often reveal mucin-like material both in the cells and in the alveolar spaces. The nuclei are usually situated at the bases of the cells, are oval and have a distinct nuclear membrane. The chromatin has a powdery appearance, and there is a single large nucleolus, which stains purple to cherry red with Masson's stain. Mitotic figures are exceedingly rare.

In older tumors the behavior of the cells is different. Their free surfaces become irregular, and it is common to see nuclei at the tips of the cells instead of at the bases. Indeed, irregularity may become so prominent that some have described "giant-cell-like masses."[22] In this phase the cells show definite proliferative tendencies, with formation of papillary processes and rare mitotic figures. The papillary proliferation may become so profuse that the limiting alveolar walls are ruptured, with coalescence of the papillary neoplastic tissue. The alveolar spaces contain free cells, which appear to be derived from the investing epithelium; however, because they are unaffected by restraining influences they are rounded. In some cases the spaces are almost filled with material which stains red with mucicarmine. If secondary infection occurs, as it frequently does, polymorphonuclear leukocytes become numerous.

The peribronchial, perivascular and pleural lymphatic channels may be lined by cells identical to those investing the alveoli, and occasionally lymphatic emboli of tumor cells are identified. At times it is possible to demonstrate a gradient from early investment of alveoli to areas of frank cancer.[11]

In the early phases the stroma of the tumor consists of normal alveolar walls. In the later lesions, especially where inflammation has occurred, there is considerable fibrosis. Squamous metaplasia is only rarely associated with fibrosis of alveolar walls and septums. Extensive endarteritic changes of the pulmonary arteries may be seen in older lesions. By the time autopsy is performed, the neoplastic proliferation has usually progressed to involve the peribronchial tissues. Careful examination will reveal that the respiratory epithelium is intact, although there may be compression of the bronchial tree by the surrounding neoplastic mass.

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21. Paul, L. W., and Ritchie, G.: Radiology **47**:334, 1946.
22. Musser, J. H.: Univ. Pennsylvania M. Bull. **16**:289, 1903.

Metastases may be present in pulmonary lymphatic channels and hilar lymph nodes and at distant sites. Their histologic appearance simulates that of adenocarcinoma; however, necrosis is almost always lacking, and the cells closely resemble those investing the alveoli.

ETIOLOGIC CONSIDERATIONS

The lungs of sheep affected with jagziekte [23] have a remarkable similarity to human lungs seen in this series (fig. 14). The same observation has been made by others.[24] Jagziekte (from *jagt,* to drive, and *Ziekte,* a sickness) has also been called verminous pneumonia,[25] epizootic adenomatosis [26a] and Montana progressive pneumonia.[27] Geographically, it has been reported in South Africa,[28] France,[29] Great Britain,[30] Iceland and Germany,[26b] the United States,[31] and Peru.[32] In 1927 Cowdry and Marsh [27] conducted an inquiry to determine whether the disease occurred in Argentina, Australia and New South Wales, but no cases resembling those of jagziekte had been reported in those sheep-raising countries.

Examination of a pulmonary lesion of a goat [33] revealed changes characteristic of jagziekte (fig. 15); however, nothing referring to caprine pulmonary adenomatosis has been found in the literature, and Geever has stated that in his experience the condition is unique.[34]

Numerous theories as to the cause of jagziekte have been proposed but have been either rejected or not substantiated. Protozoa resembling the crescent of Plasmodium falciparum were described by Robertson.[35]

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23. Case from Registry of Veterinary Pathology, Army Institute of Pathology Accession 185361, contributed by C. L. Davis, D.V.M.

24. (*a*) Sims, J. L.: Arch. Int. Med. **71**:403, 1943. (*b*) Neubuerger.[4] (*c*) Bonne.[12] (*d*) Wood and Pierson.[20]

25. M'Fadyean, J.: (*a*) J. Comp. Path. & Therap. **7**:31, 1894; (*b*) **33**:1, 1920.

26. Dungal, N.: (*a*) Proc. Roy. Soc. Med. **31**:497, 1938; (*b*) Am. J. Path. **22**:737, 1946.

27. Cowdry, E. V., and Marsh, H.: J. Exper. Med. **45**:571, 1927.

28. (*a*) Mitchell, D. T., in Third and Fourth Annual Reports of the Director of Veterinary Services, Union of South Africa, 1915, 585-614. (*b*) Cowdry, E. V.: J. Exper. Med. **42**:335, 1925. (*c*) De Kock, G., in Fifteenth Annual Report of the Director of Veterinary Services, Union of South Africa, October 1929, p. 611 and (*d*) p. 1169.

29. Aynaud, M.; Peyron, and Falchetti: Compt. rend. Acad. d. sc. **195**:342, 1932.

30. (*a*) Taylor, E. L.: Proc. Roy. Soc. Med. **31**:505, 1938. (*b*) M'Fadyean.[25]

31. (*a*) Creech, G. T., and Gochenour, W. S.: J. Agric. Research **52**:667, 1936. (*b*) Cowdry.[26b] (*c*) Cowdry and Marsh.[27]

32. Caparo, A. C.: Bol. escuel. van de cienc. vet. **1**:27, 1945.

33. Case from Registry of Veterinary Pathology, Army Institute of Pathology Accession 185359, contributed by C. L. Davis, D.V.M.

34. Geever, E. F.: Personal communication to the author.

35. Robertson, W. J.: Comp. Path. & Therap. **17**:221, 1904.

Cowdry [25b] was unable to demonstrate spirochetes. M'Fadyean [25a] suggested that in Great Britain verminous pneumonia was caused by a nematode, Strongylus rufescens; however, the common lung worm, especially in Iceland,[26b] is Muellerius capillaris. It is the consensus that these nematodes are not the etiologic agent, but they may be a predisposing factor.[25b]

Pulmonary growths resembling jazgiekte have been encountered in numerous species of animals as well as in man. Grumbach [36] described alveolar epithelial hyperplasia in guinea pigs following injection of diphtheroid organisms. Cowdry [27] had the opportunity of comparing Grumbach's slides with those from cases of jagziekte and was able to verify their close similarity. Jagziekte of horses and mules was described by Theiler.[37] Experimental contact, injections of emulsified organs and pericardial fluid, and drenching in urine did not convey the disease to healthy horses and mules. He conclusively proved that the equine disease was caused by a plant, Crotalaria dura. Although his microscopic descriptions and photographs do not entirely convince one that the changes are identical with those seen in sheep affected with jagziekte, his work is remarkable in that he has presented adequate evidence that a noxious plant as well as specific micro-organisms can produce a typical pulmonary disease. De Kock,[25c, d] in experiments conducted at Onderstepoort and Tweespruit, South Africa, was unable to reproduce jagziekte in sheep with contact. He states that it can safely be said that jagziekte does not belong to the category of ordinary infectious diseases.

Olafson and Monlux [38] examined a cat infected with Toxoplasma, and found the alveoli of the lung lined by large epithelial cells, which gave the affected portions an adenomatous appearance. The alveolar walls were thickened and hyperemic.

Tyzzer [39] and others [40] described "papillary cystadenoma" occurring in the lungs of mice. The observation has since been made that such tumors are exceptionally common in this species, although uncommon in other mammals with the exception of sheep.

It has been shown experimentally that carcinogenic hydrocarbons can produce pulmonary tumors in mice. The pulmonary lesions of mice described recently by Lorenz and Stewart [41] closely resemble the human

36. Grumbach, A.: Bull. Assoc. franç. p. l'étude du cancer **15**:213, 1926.

37. Theiler, A., in Seventh and Eighth Reports of the Director of Veterinary Research Department of Agriculture, Union of South Africa, 1918, p. 59.

38. Olafson, P., and Monlux, W. S.: Cornell Vet. **32**:176, 1942.

39. Tyzzer, E. E.: J. M. Research **21**:479, 1909.

40. (*a*) Slye, M.; Holmes, H. F., and Wells, H. G.: J. M. Research **25**:417, 1914. (*b*) Wells, H. G.; Slye, M., and Holmes, H. F.: Cancer Research **1**:259, 1941. (*c*) Grady, H. G., and Stewart, H. L.: Am. J. Path. **16**:417, 1940.

41. Lorenz, G., and Stewart, H. L.: J. Nat. Cancer Inst. **7**:227, 1947.

type of adenomatosis. On the other hand, the tumors reported by Grady and Stewart[40c] were actually papillary adenomas and distinct from pulmonary adenomatosis of man.

Dungal,[26b] whose experimental work was carried on in Iceland where jagziekte is a serious problem, found that the infection was readily brought about when healthy lambs were exposed to the exhaled breath of sick sheep. He was unsuccessful in reproducing jagziekte by introducing bronchial secretions or ground tissues from affected sheep into the lungs of healthy animals. However, if sheep were already infected with lung worms it was possible to produce the lesions of jagziekte with extracts of diseased tissue, if this was given in conjunction with intratracheal injections of cultures of bacteria that cause pneumonia in sheep. In spite of the prevalence of jagziekte in the island, Dungal stated that he had never seen a similar lesion in the human lung in the numerous autopsies which he had performed. This observation is in disagreement with those of other investigators, who have described lesions of the human lung closely resembling those of jagziekte.

Experimental transmission was attempted with material from 2 human subjects,[42] but the results obtained with that of one are alone available.[24a] The pulmonary tissue of this one injected into rabbits, guinea pigs and monkeys had no effect. It should be noted that sheep were not included, and the species used, with the possible exception of guinea pigs, probably have low tissue susceptibility to this condition.

To date there is nothing in the literature to indicate that human beings contract jagziekte from animals, and an infectious origin cannot be determined from the evidence available.[43]

HISTOGENESIS

The controversy regarding the genesis of this tumor traverses the gamut from claims for a particular cell of origin [44] to the denial of the very existence of such a tumor entity.[45] The recent literature, however, has in the main shown a significant trend toward acceptance of the theory of an alveolar epithelial origin.[46]

If one is to assume that the tumor arises in the peripheral portion of the lung, it would be logical to consider the evidence for the presence

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42. Wood and Pierson.[20] Sims.[24a]

43. Dungal.[26b] Creech and Gochenour.[31] Theiler.[37]

44. Neubuerger and Geever.[1] Neubuerger.[4]

45. Herbut, P. A.: (*a*) Am. J. Path. **20**:911, 1944; (*b*) Arch. Path. **41**:175, 1946. (*c*) Arkin and Wagner.[17]

46. (*a*) Ikeda, K.: Am. J. Clin. Path. **15**:50, 1945. (*b*) Smith, L. W., and Gault, E. S.: Essentials of Pathology, ed. 2, New York, D. Appleton-Century Company, Inc., 1942. (*c*) Wenger, F.: Prensa méd. argent. **32**:44, 1945. (*d*) Taft, E. B., and Nickerson, D. A.: Am. J. Path. **20**:395, 1944. (*e*) Bell.[13]

or the absence of an alveolar lining. The work of investigators has yielded diametrically opposed results, as evidenced in the comprehensive reviews of Miller [47] and Loosli.[48] Miller recognized that the greatest obstacle to an understanding of alveolar epithelium was the impossibility of dissecting it off. However, in certain pathologic processes, for example, the outpouring of a serous exudate behind the epithelium, as in pneumonia, atelectasis [49] and pulmonary edema of mitral stenosis, he observed that the fluid peeled the epithelium from the alveolar walls. He concluded that the lining of the alveolar walls was a continuous epithelium. This mechanism of vis a tergo had also been described in 1936 by Gazayerli,[50] who found that certain substances when injected into the pleural sac rendered the alveolar lining visible just as disease processes do. Cooper [51] studied the histologic evidence presented in human material and concluded that there is a fetal alveolar epithelium which persists but becomes attenuated when air distends the lungs. This same observation has been made by Bensley and Groff [52] and by Zeldes.[53]

Bremer [54] believed that the alveoli were lined with a continuous layer of epithelium of entodermal origin and that the cytoplasm of the component cells extended over the alveolar capillaries with flangelike processes.

Palmer and others [54] were of the opinion that there was a discontinuous lining epithelium of the alveolus, and Sprunt,[54] that there were two kinds of cells: one a phagocytic mesenchymal cell, the other an epithelial lining cell.

Loosli [48] and others [55] were unable to find irrefutable evidence of the existence of an epithelial lining. Macklin [56] observed that because a lining epithelium could not be demonstrated, the number of those denying its presence had increased. He recognized that if this opinion were generally adopted one would have to reevaluate the current concepts of the alveolus so as to regard it as "a functional interstitial emphysema." According to Cooper,[51] "these views are incompatible with histological, pathological and embryological evidence, and the arrangement of free spaces communicating with the exterior and in

47. Miller, W. S.: The Lung, ed. 2, Springfield, Ill., Charles C Thomas, Publisher, 1947.

48. Loosli, C.: Am. J. Anat. **62**:375, 1938.

49. Löhlein, M.: Verhandl. d. deutsch. path. Gesellsch. **12**:111, 1908.

50. Gazayerli, M. E.: J. Path. & Bact. **43**:357, 1936.

51. Cooper, E. R. A.: J. Path. & Bact. **47**:105, 1938.

52. Bensley, S. H., and Groff, M. B.: Anat. Rec. **64**:27, 1935.

53. Zeldes, M.: Arch. Path. **20**:748, 1940.

54. Cited by Macklin.[56]

55. (*a*) Lang, F. J.: Virchows Arch. f. path. Anat. **275**:104, 1930. (*b*) Norris, R. F.; Kochenderfer, T. T., and Tyson, R. M.: Am. J. Dis. Child. **61**:933, 1941.

56. Macklin, C. C.: J. Thoracic Surg. **6**:82, 1936.

contact with thin-walled capillaries does not accord with histological findings in general."

Bloom [54] expressed the belief that mesenchymal cells may assume an epithelial arrangement and that the lining separated from the alveolar wall in pathologic conditions may be composed of mesodermal cells and exudate. Fried [57] regarded cells found in alveoli as mesodermal phagocytes originating in the alveolar septums. Lang [55a] introduced the term "septal cell" to indicate a special phagocyte belonging to the reticuloendothelial system, equivalent to the epicyte (Clara). Macklin [56] recognized the possibility that these cells may become the nucleus of a primary pulmonary carcinoma. Neubuerger and Geever [1] proposed that the so-called alveolar cell tumor was derived from septal cells. It is well to note that Miller was unconvinced that this cell was anything more than an altered epithelial lining cell, or at most a mononuclear leukocyte, and Herbut [45a] was unable to find any convincing evidence that the septal cell was the parent cell of the tumor cell.

Dungal [26b] inclined to the view that in jagziekte the mononuclear exudate cells were derived from the alveolar lining and that the lining cells could proliferate, without losing contact with the alveolar wall, to form an epithelial tuft which might be the beginning of an epithelial growth. De Kock [28c] concluded from the results of his studies of sheep that the majority of proliferations arose from alveolar epithelium.

Studies in mice have been numerous, and Furth and Furth,[58] Grady and Stewart [40c] and others [59] were of the opinion that the papillary tumors arose in the alveoli.

RELATION OF PULMONARY ADENOMATOSIS AND CARCINOMA

It is a misconception that regional lymph node or generalized metastases are never present with pulmonary adenomatosis. However, Richardson [8] was of the opinion that these tumors represented a distinct type of epithelial growth without formation of metastases. Ikeda [40a] considered only one of the tumors he reported a typical example of what he called alveolar cell carcinoma of the lung, for, among other criteria, there were no regional lymph node or distant metastases. Paul and Ritchie [21] discussed a "benign" and a "malignant" form of pulmonary adenomatosis.

The case reports of Malassez [60] and Musser [22] are commonly accepted as the first to present the nodular and the diffuse type, respectively, and in both regional lymph node metastases were described.

57. Fried, B. M.: Arch. Path. **17**:76, 1934; cited by Macklin.[56]

58. Furth, J., and Furth, O. B.: Am. J. Cancer **34**:169, 1938.

59. McDonald, S., Jr., and Woodhouse, D. L.: J. Path. & Bact. **54**:1, 1942. Tyzzer.[39] Slye and others.[40a] Wells and others.[40b]

60. Malassez, L.: Arch. de physiol. norm. et path. **3**:353, 1876.

It is the accepted procedure to base criteria for any condition on the original description. If time and experience demonstrate errors, then new or modified criteria are accepted. However, there has been no evidence presented in seventy-one years which alters in any way the original descriptions; therefore, they may be considered classic.

In 1907 Helly [2] discussed the possibility that the pulmonary lesion which he observed in a 43 year old woman may have been cancer. In 1930 Oberndorfer [62] stressed the fact that there is no distinct borderline between alveolar investment, which histologically appears benign, and frank carcinoma of the lung. He concluded that in the lung, as in the parenchymatous cells of the liver, sudden mutations of the epithelium may arise in several areas leading to "blastoma" formation.

Bonne,[12] in 1939, suggested the term carcinosis for the pulmonary lesion under discussion. He was puzzled by the histologically "benign" appearance of the tumor and the contradictory quality of "malignancy" represented by invasion of pleura and death of the patient. Dacie and Hoyle,[11] in reporting a case in 1942, illustrated "a fine gradation histologically from benign 'adenomatosis' to definite invasive carcinoma." The same year Smith and Gault [46b] included in their textbook an account of a case in which they described innumerable mitotic figures and stated, ". . . some suggestion of lumen formation is seen indicating the possibility that the cells are of actual alveolar lining cell origin with a tendency toward adenocarcinomatous formation." In 1943 Bell [18] wrote: "There is no good reason to doubt that hyperplasia of the alveolar epithelium may give rise to localized or diffuse growths which may form metastases." In 1944 Geever, Carter, Neubuerger and Schmidt [14b] commented that the majority of "alveolar-cell tumors are malignant." Finally, Paul and Ritchie [21] concluded from evidence in one of their cases that "adenomatosis" must be regarded as a precancerous lesion.

Comparative Pathology.—Aynaud, Peyron and Falchetti,[29] who examined numerous cases of verminous pneumonia originating in France, concluded that the process was a real tumor. Dungal had the opportunity of examining the sections of jagziekte studied by this group and stated "there were undoubted metastases in a lymph gland." Innes [63] was also convinced that jagziekte was neoplastic in nature, and De Kock,[28c] that serious consideration should be given its "malignant" potentialities. De Kock and others [40c] pointed out the similarity between jagziekte and spontaneous pulmonary tumors of mice. In 1911

61. Footnote deleted by the author.

62. Oberndorfer, S.: Virchows Arch. f. path. Anat. **275**:728, 1930.

63. Innes, J. R. M., in discussion on Taylor.[30a]

Haaland [64] examined a large series of primary pulmonary tumors of mice and concluded that they were "malignant."

Numerous investigators have induced pulmonary tumors in mice with a variety of irritants.[65] Campbell [66] observed primary pulmonary tumors in mice exposed to dusts and tars of various kinds and was of the opinion that they usually originated from alveolar cells as "non-malignant" tumors and then changed more or less rapidly into "malignant" types, depending on the degree of irritation.

COMMENT

General Considerations.—The lesions seen in the lungs of sheep, goats, horses and mules, guinea pigs, cats and man must be interpreted with considerable caution. Numerous investigators have discussed the striking similarity between jagziekte and alveolar cell tumor of the human lung. I have had the opportunity to compare sections from certain spontaneous and induced pulmonary tumors in mice [41] with those of goat, sheep and man, and it is apparent that the pulmonary lesions closely resemble one another. Therefore, it is possible that an identical pulmonary reaction takes place in these widely divergent species, and a review of the causes of adenomatosis in these species may shed light on the cause of this condition in man.

Etiologic Factors.—It seems established that widely assorted irritants acting on the lung must produce a rather stereotyped response regardless of species; i. e., investment of the alveolar walls. The stimulating agents may be exogenous and include chemical fumes or other respiratory irritants, or they may be endogenous and include the extrarespiratorily introduced carcinogens, ingestion of certain poisonous weeds, bacteria, protozoa and possibly viruses. There is evidence that investment of alveoli occurs in the human lung in a wide assortment of chronic pulmonary conditions, such as pneumonia, tuberculosis, atelectasis, psittacosis and chronic passive congestion. No etiologic factor has been proved to be specific. If a virus or viruses should be isolated, they would probably be either contributory or another one of the innumerable irritants. The possibility does exist that diverse irritants may prepare the "ground" for the action of a virus, but from the evidence available this seems unlikely.

Histogenesis.—The origin of the epithelium-like tumor cells in pulmonary adenomatosis is still undetermined. However, the presumptive

64. Haaland, M.: Fourth Scientific Report of the Imperial Cancer Research Fund, London, Taylor and Francis, 1911; Proc. Roy. Soc. Med. **83**:520, 1911.

65. Andervont, H. B., and Shimkin, M. B.: J. Nat. Cancer Inst., **1**:225, 1940. Leiter, J.; Shimkin, M. B., and Shear, M. J.: ibid. **3**:155, 1942. Grumbach.[86] Grady and Stewart.[40e] Furth and Furth.[58]

66. Campbell, J. A.: Brit. J. Exper. Path. **18**:215, 1937.

evidence obtained from clinical, experimental and pathologic data leads me to conclude that a continuous alveolar lining probably exists.

In no instance could the available evidence be interpreted as indicating that the alveolar investing cells arose from bronchi by a process of extension, as Herbut claimed. It is noteworthy that in 1 case the investing cells were found to be ciliated. This is an unusual finding, reported once before.[21] There is no reason to believe that these cells arose in the bronchi; ciliation is merely an expression of the pluripotential character of the parent cell. It has also been shown that the investing cells may be predominantly mucus-secreting and may look and act like goblet cells.

It is obvious that ciliated or goblet cells are not autochthonous to the pulmonary alveolar walls. As long as proof is lacking that they have their origin from the bronchial epithelium, it is my opinion that the alveoli contain some cells capable of attaining autonomy.

Pathogenesis.—Comparison of material from human and experimental sources leads to the deduction that the cells which line the alveoli in many pathologic conditions have the same origin as the cells which are present in the so-called alveolar cell tumor. It is common knowledge that if the cause of pulmonary irritation is withdrawn, the inflammation may resolve completely. If the irritation continues, the first changes are seen in the respiratory epithelium and the bronchial glands. Within the glands there is an increase in the number of goblet cells and apparent hyperplasia of the mucous glands, and a considerable quantity of glairy mucus exudes over the respiratory epithelium and acts as a protective coating. This is manifested clinically by increasingly abundant mucoid sputum.

Up to this point there is little if any disagreement among investigators. The following conclusions have been evolved by study of the monotonous repetition of the histologic picture in all chronic pneumonitides. If the irritation continues, a remarkable change takes place in the alveoli themselves, characterized by the appearance of alveolar investing cells. The investing lining cells are, in all probability, a recapitulation of the protective function displayed in the bronchi, already described.

It becomes apparent at this phase that the hyperplasia of the alveolar lining cells has reached a stage of "new growth," where it ceases to serve a useful function for the organism. It has been demonstrated that continued proliferation of the lining cells invests extensive areas of lung parenchyma and obviously acts as a barrier to normal physiologic function of the lungs. Thus the patient may literally "drown" in tumor tissue before the true cancerous propensity of the proliferation is made obvious by distant metastases, as in case 9.

SUMMARY

"Pulmonary adenomatosis" is suggested as the preferable designation for so-called alveolar cell tumor of the human lung. If metastases occur, the term should be modified by adding the adjective "cancerous" (cancerous pulmonary adenomatosis).

Criteria proposed for the diagnosis of pulmonary adenomatosis are: (1) alveolar cellular proliferation characterized by the appearance of tall columnar mucus-producing cells; (2) absence of an intrinsic tumor of the bronchial tree, and (3) absence of primary adenocarcinoma of any other part of the body.

No etiologic factor of pulmonary adenomatosis of man has been proved to be specific; the disease apparently is not infectious.

The majority of observers are of the opinion that pulmonary adenomatosis is an extrabronchial neoplasm with cancerous potentialities. In none of the cases presented was there conclusive evidence as to the exact site of origin.

Although these tumors appear histologically noncancerous, clinically they must be considered to be cancers, since they may kill by local growth or by metastasis.

THE MEGAKARYOCYTE

III. Pseudothrombocytes

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AS MAY be seen from Hittmair's [1] extensive review, there is no constituent of the hemopoietic tissue which was not at one time or another considered to produce thrombocytes. The interpretation of these structures varied from a complete cell to a mere precipitate. The morphologic features and distinctive reactions of the thrombocytes point to unity of function, form and origin. At present the megakaryocyte is almost unanimously accepted as their only source. Nevertheless the unity of their formative process is not generally recognized. Whereas the original theory of Wright assumes that the thrombocytes are liberated by disintegration of the mature, granulated megakaryocyte, other workers [2] have ascribed thrombocytopoietic activity also to the early, nongranulated stages. At this early phase pseudopodia are said to protrude, which when separated from the cell body either represent basophilic juvenile thrombocytes [2e] or produce a chromomere from nuclear chromatin particles which have been dislocated into them.[2b, d] These assumptions imply two significantly different processes: (1) the liberating of thrombocytes from the mature megakaryocyte, after completion of granulopoiesis, and (2) the segregating of the hyalomere from the basophilic cell, before the start of granulopoiesis, followed by secondary elaboration of the chromomere. Although Willi [2b] and Rohr and Koller [2d] assumed that the nuclear chromatin participates in both mechanisms, the succession of the formative steps remains inversed. The secondary production of granules in the pseudopodia, furthermore, is not in accordance with the granulopoiesis that occurs in the megakaryocyte described by me.[3] Granulopoiesis must be considered as a monocentric process, starting in the "functional area," which gradually expands over the cell body without any intervention of

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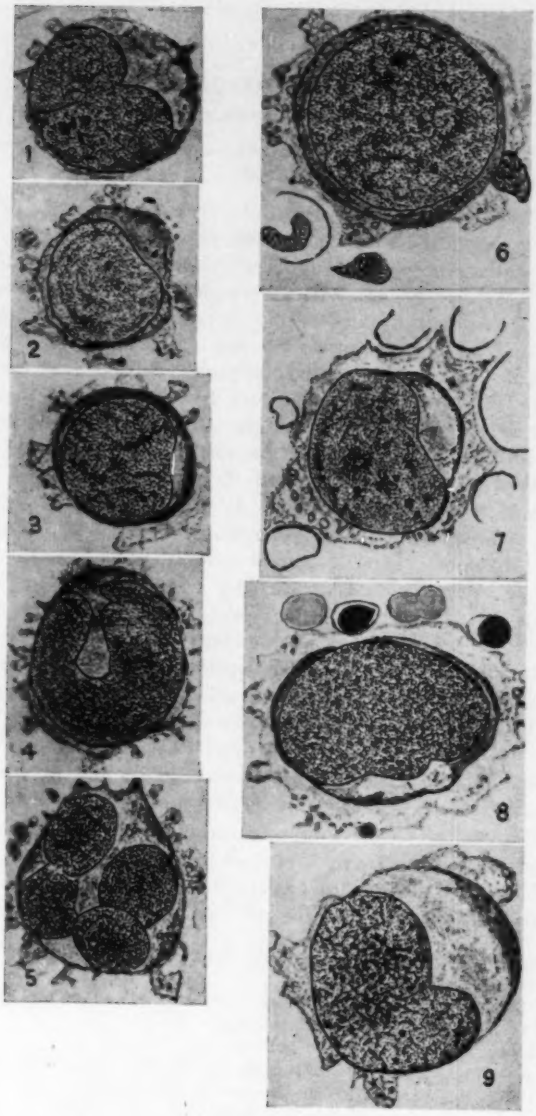
The Department is supported in part by the Michael Reese Research Foundation and the Hematology Research Foundation.

1. Hittmair, A.: Folia haemat. **35**:156, 1927.

2. (*a*) Medlar, E. M.: Folia haemat. **53**:397, 1935. (*b*) Willi, H.: ibid. **53**:426, 1935. (*c*) Juergens, R., and Graupner, H.: ibid. **57**:263, 1937. (*d*) Rohr, K., and Koller, F.: Klin. Wchnschr. **15**:49, 1936. (*e*) DuBois, A. H.: ibid. **16**: 22, 1937. (*f*) Downey, N., and Nordland, U.: Folia haemat. **62**: 1, 1939.

3. Schwarz, E.: (*a*) Arch. Path. **45**:333 and (*b*) 342, 1948.



Figures 1-9

(See legends on opposite page)

accessory granulopoietic foci or chromatin particles. For these reasons the aforementioned findings should permit another interpretation, which necessarily will reflect also on the pathologic changes of the thrombocytes. The following observations may contribute to the clarification of these controversial issues.

OBSERVATIONS

The studies were made on smears and sections of human marrow, which was obtained by sternal puncture from persons with either normal or morbid hemopoiesis. The usual staining methods were applied.

Distribution of "Pseudopodia."—Threadlike, fringed or lobiform excrescences are a common deformity of early megakaryocytes. Such "pseudopodia" appear on almost all the megakaryoblasts, are less readily observed on the promegakaryocytes and are rarely seen on the mature cells. The frequency or intensity of the fimbriation has no relation to the general condition of hemopoiesis or the number of thrombocytes. Numerous instances of fimbriation were observed also in thrombopenic purpura.

Morphologic Aspects.—1. The number of excrescences varies from a single one to a wreath or corona of them (figs. 1 to 5). Some may appear as a filiform fringe (fig. 4), or may display clublike ends (figs. 2 and 3), or may show quite irregular shapes, but every one of them has a narrow insertion on the surface of the cell. Other protrusions with a broader base are more or less lobiform (figs. 1, 3, 6 and 7). Instead of pseudopodia, apposition-like formations frequently stretch along the cell outline, either replacing the lobes or being in continuity with them (figs. 3, 6 and 9). These structures show an indefinite outline, are usually interrupted, and rarely and only in very immature cells do they form a continuous envelope around the cell (figs. 7 and 8).

Fig. 1.—Megakaryoblast with few excrescences.

Fig. 2.—Megakaryoblast with many pseudopodia-like excrescences, one of them detached and simulating a basophilic thrombocyte.

Fig. 3.—Megakaryoblast with many excrescences and some continuous appositions.

Fig. 4.—Early promegakaryocyte with a corona of excrescences.

Fig. 5.—Promegakaryocyte with many excrescences, some of them detached, the latter representing pseudothrombocytes.

Fig. 6.—Promegakaryocyte with a nearly continuous envelope. Note the very large single nucleus resulting from a monocentric mitosis.

Fig. 7 and 8.—Early megakaryoblasts, each with a continuous envelope and a small functional area.

Fig. 9.—Maturing promegakaryocyte with a large functional area and a few lobiform excrescences.

Note the sharp line of demarcation between the cell body and the pseudopodia in all figures. Magnification about 1,000.

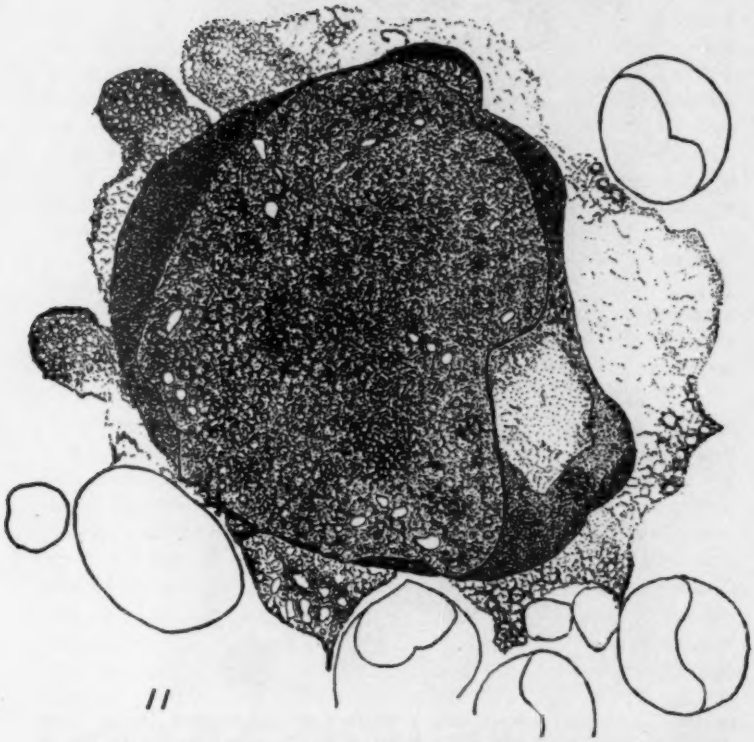
As far as they contain solid material the protrusions stain in various shades of blue, but always they are lighter than the cytoplasm. A sharp line of demarcation separates the bases of the protrusions from the cell body. In places where the denuded nucleus occupies a part of the cell contour, close observation reveals a thin blue line separating the bases of the protrusions from the nuclear membrane (figs. 1, 4 and 5).

2. One type of excrescence shows basophilic threads and granules embedded in an almost colorless ground substance. The filaments form a loose and irregular meshwork and are composed of minute dotlike or beadlike granules, which at the point of intersection are often coarser and more conspicuously stained. In another type the more homogeneous ground substance stains distinctly blue and is perforated by numerous small holes varying from pinpoint size upward, producing a foamy appearance (figs. 3 and 6). The holes, usually circular, contain no material visible either by stain or by refraction of light. The surface of the slide provides the background of these absolutely empty spaces. In order to show the structural differences, which are rather difficult to reproduce in drawings, figures 10 and 11 were drawn on the double scale. Not only the structural properties but the sharp line of demarcation and the well defined "functional area" are worth noticing.

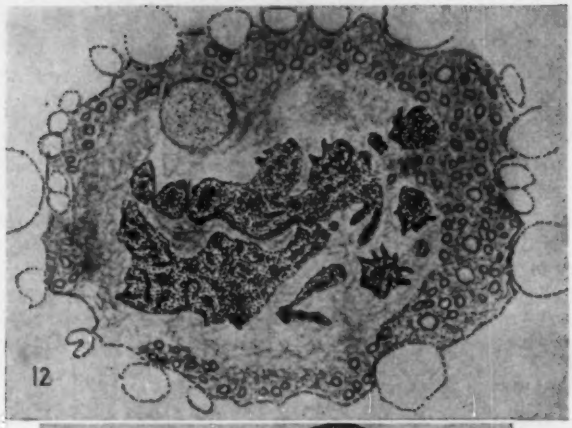
In another configuration the foamy structure occupies either larger lobes [4] or a zone of varying breadth of the cytoplasm. In both cases the line of demarcation is absent. In figure 12, representing a promegakaryocyte in pluripolar mitosis, the dark foamy zone surrounds the central homogeneous and pale chromosomal field.

3. In many fields of the films, in places where the cells have not spread too far from each other the dried sap between the marrow cells shows small holes just the same as those described. If the cells are widely separated, the intercellular material is almost invisible. The holes are absent where the cells are tightly crowded. Variations in the thickness of the film and in the blood plasma dilution of the sap account for these differences. In the preparation of the smear the slides do not move either in the same plane or with constant velocity, and therefore pressure and suction alternate and drive air into the sap. These air bubbles are not retained in the thinner fluid layers but remain enclosed in the thicker and more viscous ones, thus forming a true foam. In the drying process the bubbles explode, leaving the round perforating holes. Moreover, the various physical conditions cause irregularities of the surface tension, a factor which probably is responsible for the filaments and the granular precipitations in the drying sap.

Applying this interpretation to the identical foamy appearance of many protrusions and parts of the cytoplasm of the megakaryocytes,



Figs. 10 and 11.—Megakaryoblasts with initial functional area. Note the many lobes, pseudopodia and some continuous appositions. The foamy structure and the continuous transitions from one type of excrescence to another are very clear. Note the distinct line of demarcation. The cells are drawn on a double scale in order to show the structural details. Magnification, about 1,000.



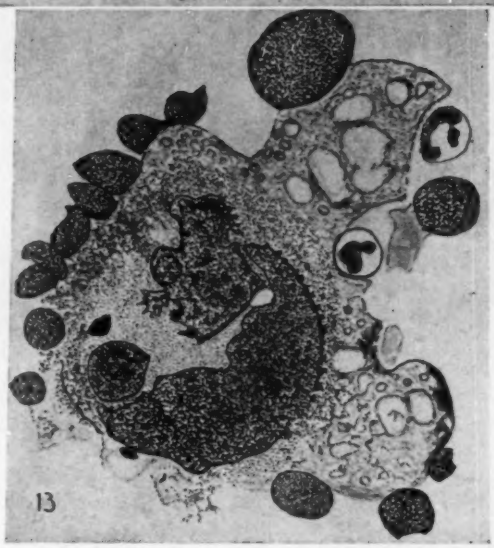


Fig. 12.—Megakaryoblast with a smashed pluripolar mitotic figure. The peripheral zone of the cell shows foamy structure. There is here no line of demarcation. The circular functional area is distinctly outlined. Magnification about 1,000.

Fig. 13.—Mutilated mature megakaryocyte. Note the large lobes with holes and foamy structure. Here there is no line of demarcation. Magnification, about 1,000.

one perceives that these features do not represent a genuine structure or vacuolation. They are the result of pressure and stretching and of the viscosity of the material on which these forces act.

Obviously, the peripheral parts of the cell are foremost in yielding to these forces, because of the decrease of limiting obstacles toward the exterior. From these points of view the features of the cell drawn in figure 12 are easily understood, as well as the origin of the structures described as pseudopodia and lobiform protrusions. Futhermore, the presence or the absence of the demarcation line decides whether the lobes belong to a substance outside the cell surface or to the cell body itself, a distinction which will prove itself of principal significance.

The movements of the slides develop not only pressing and stretching but also shearing forces which lacerate the material and tear off more or less of it, thus forming the pseudopodia-like excrescences and the thrombocyte-like isolated corpuscles. Lobes with a broader base result in places where the forces are weaker. The appositions described are but the residues of the flattened material spared from the shearing off and from the laceration. Their lack of the foamy appearance suggests a lesser viscosity and consequently a stretching into thinner layers. That there are local variations of all these physical factors is rendered evident by the fact that there are transitions from one structural type to the other (figs. 10 and 11).

The protrusions, decreasing in number with progressing maturation, become gradually reduced to one or two foamy lobes, usually combined with serious mutilation of the cells (fig. 13). Sometimes the peripheral ungranulated zone of the mature megakaryocyte shows the foamy transformation. In such instances the line of demarcation is absent (fig. 13).

Participation of the Nucleus.—Chromatin particles entering into the pseudopodia and suggesting that they share in the formation of the chromomere were never encountered. Even within the main cell body such particles are extremely rare, although the pluripolar mitosis so characteristic of the megakaryocyte would favor such occurrence. In a few instances (fig. 14) budlike excrescences of the nuclei were limited to seriously injured cells. In figure 17, which is similar to the megakaryocyte depicted by Willi[2b] (fig. 5 in his paper), the discontinuities and the serrated contour of the nuclear membrane suggest that the isolated extranuclear chromatin crumbs originated through mechanical dislocation. In all similar instances (figs. 14, 15 and 16) the particles were never found within a protrusion and never showed any sign of further transformation. This apparent inactivity of the particles fits well with the behavior of an artificial product.

Relations of Pseudopodia and True Thrombocytes.—Of all the protrusions, only those with clublike ends show greater resemblances

are produced in these sites. The correct interpretation of such misleading pictures presumes a detailed inquiry into the manner in which thrombocytes are liberated from the mature megakaryocytes and also into the possibility that one could be deluded by thrombocytes superimposed on, or apposed to, immature cells. These will be reported in another paper. Only a few points will be considered in the following comment.

COMMENT

From these observations the following significant facts emerge:

1. A distinct line of demarcation separates the bases of the excrescences from the main cell body.

2. The shape and the size of excrescences vary from filaments to lobes, and the number from a single protrusion to a corona of protrusions and eventually to a continuous envelope of the early megakaryocyte.

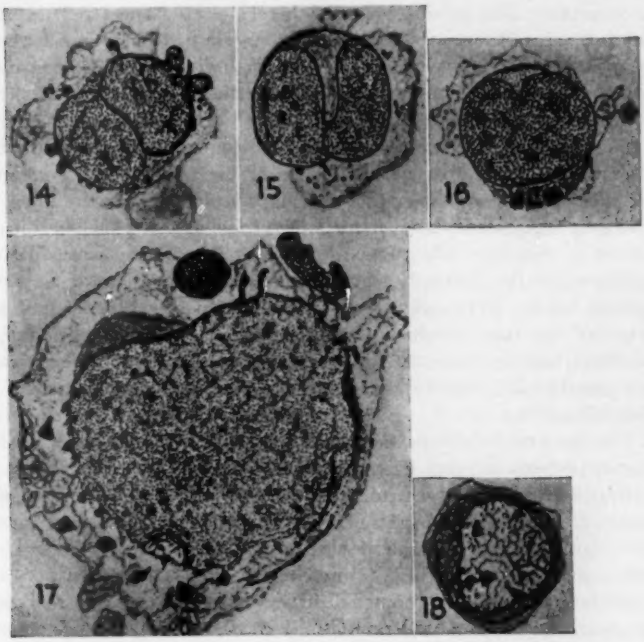
3. The protrusions exhibit structures identical with those seen in the intercellular sap of the smear, where they are formed according to the local variations of mechanical forces, the thickness of the film and the viscosity of the material.

4. Neither chromatin particles nor granules which do not stem from the functional area of granulopoiesis of the main cell body appear in the protrusions.

Consequently the existence of the line of demarcation does not permit one to interpret the excrescences as pseudopodia, since the latter as a product of ameboid activity obviously should stand in direct continuity with the cytoplasm of the cell body. This statement, together with the observations concerning the shape and the structural properties of the excrescences, qualifies them as the locally variable effects of the spreading and shearing forces acting on a viscous material that surrounds the megakaryocytes in their early stages. Therefore, the detached basophilic bodies carrying the features of the protrusions cannot be valued as the products of a vital development. They are artificially formed by the same forces as the protrusions themselves, without any relation to true thrombocytes.

The study of artefacts, though usually a sterile affair, yields here an interesting conclusion. An artefact produced by physical and chemical agencies is either inevitable in or accessory to the technical procedures to which the studied substrate was submitted. The constant occurrence of a special artefact stipulates that it must be related to certain properties of the substrate itself. The protrusions studied here appear on megakaryocytes of early developmental stages only and correspond to an artificial stretching and lacerating of a continuous layer. Therefore, this envelope represents a general property and either may be

to thrombocytes in their shape. They differ widely from the latter, however, in their texture. The possibility of a wrong interpretation is greater with the detached bodies; they show the structure of the protrusions, sometimes even their foamy appearance, but not the characteristics of the hyalomere of true thrombocytes. Granules in their interior stain basophilic (figs. 2 and 3) but do not show the azure-like tint of a chromomere. Obviously, larger excrescences, especially the lobes, do not have to be considered.



Figs. 14 to 16.—Megakaryoblasts showing nuclear buds and dislocated chromatin particles.

Fig. 17.—Promegakaryocyte. The nuclear membrane has been artificially destroyed at some places. Note the dislocated chromatin particles in the cytoplasm.

Fig. 18.—Early megakaryoblast with a narrow layer which enwraps nearly the entire cell body. From a section of marrow fixed in Zenker-formaldehyde solution; Giemsa stain.

All the cells in figures 13 to 17 show clearly the mechanical deformation. Magnification, about 1,000.

Unmistakable thrombocytes appearing between the protrusions or situated in the vicinity of, or on, the body of such a megakaryocyte probably are primarily responsible for the assumption that thrombocytes

itself of an artificial origin or may be a preformed though artificially deformed structure of the early megakaryocyte. The first hypothesis presumes that the mechanical forces expel from the cytoplasm a more fluid material, which thereafter is submitted to deformation. It is difficult to imagine that pressure and stretching may act so regularly on a cell of the size of a megakaryoblast and in such a way as to cause some material to be expelled in all directions around the cell. Neither would this hypothetic origin explain the constant and unbroken line of demarcation between the cell body and the corona of protrusions. The assumption that there is a preformed thin layer of a substance of lesser viscosity around the cell, stretched and disrupted by pressure and shearing, fits much better with all the manifest features of the protrusions and especially with the line of demarcation. Demonstration of the undisturbed layer in sections may permit a definite decision. However, as mentioned previously in these studies,[3a] the preserved three dimensional shape of the cell, the shrinking with fixation and embedding and the great difficulty of differentiating the youngest megakaryoblasts from other cells are responsible for the failure of this approach. Only after a long search may a cell like that in figure 18 be observed, suggesting the existence of such an extracellular layer. This layer should not be confused with the most outward of the three zones of cytoplasm of the mature megakaryocyte distinguished by Heidenhain[5] and confirmed by Frey.[6] These zones are intracellular, whereas the layer in question is superimposed on the cell contour.

In the first communication of this series[3a] it was pointed out that the hypothesis concerning a nuclear origin of the megakaryocytic granulation does not seem to be sufficiently substantiated. The statement of the artificial nature of the protrusions eliminates automatically any significance of chromatin particles which accidentally may have been dislocated into the excrescences. Thus any suggestion that thrombocytopoiesis has occurred in the ungranulated megakaryocyte becomes untenable. One can also dismiss the singular assertion of Hadorn[7] that the whole nucleus disintegrates into thrombocytes in the purpura caused by sedormid[®] (allylisopropylacetylcarbamide).

The observation that true thrombocytes are present in the vicinity of the protrusions or apparently within some of them or in the basophilic cell body itself accounts for the assumption of a precocious activity of these cells. As early as 1923 Naegeli[8] interpreted the

4. Schwarz, [3a] figure 1*H*.
5. Heidenhain, M.: Arch. f. mikr. Anat. **43**:423, 1894.
6. Frey, H. C.: Frankfurt. Ztschr. f. Path. **36**: 419, 1928.
7. Hadorn, W.: Schweiz. med. Wchnschr. **66**:1273, 1936.
8. Naegeli, O.: Blutkrankheiten und Blutdiagnostik, Berlin, Julius Springer, 1823.

accumulation of thrombocytes around nuclear fragments in the blood of patients with chronic myelosis as selective agglutination. This much discussed matter was settled by Undritz and Röthlin.[9] These authors compared smears prepared immediately and after a delay of one-half to one minute. Thrombocytes and nuclear fragments were well separated in the first smears but frequently found in aggregation in the later ones. Smears of rabbit marrow taken with and without citrate showed similar differences. Hemmeler[10] examined the marrow of patients with severe thrombopenic purpura with and without addition of normal blood. The admixed thrombocytes were found to gather frequently around the megakaryocytes, while in the other sample only single thrombocytes were seen to be attached to them. That thrombocytes have a capacity for selective agglutination is a well established fact.

My own extensive observations harmonize with the interpretation that superposition and apposition account for most of the deluding pictures. This is the more valid for figures 15 and 16 of Medlar[2a] and figure 4 of Juergens and Graupner,[2c] in which these strands of thrombocytes seem to emerge from the basophilic megakaryocytes with a smooth contour. Nevertheless, the observation of massive accumulation of thrombocytes around free megakaryocytic nuclei does not agree entirely with this explanation. A final decision may depend on whether the thrombocytes are liberated from the mature megakaryocyte singly or in strands at the periphery of the cell or whether they are set free by simultaneous disintegration of the entire cytoplasm. This hypothetic "explosive" disintegration was first advanced by Rohr and Koller[2d] as an additional mechanism besides the partial liberation of thrombocytes. It may, however, be emphasized that Sabin's[11] studies using the technic of supravital staining support the total disintegration as the more probable and regular process.

The elimination of the pseudopodia-like protrusions of the early megakaryocytes from any participation in the production of thrombocytes restores the notion of the unity of that process. Furthermore, it contradicts the developmental system of Juergens and Graupner,[2c] because no other way is known which would furnish thrombocyte-like bodies without a chromomere, assumed by these authors to be the juvenile stages. There is nothing else described in the literature or encountered in any of my observations which could be interpreted as a segregation of basophilic thrombocyte-like bodies lacking granulation. The arrangement of the granules of the megakaryocyte in the

9. Undritz, E., and Röthlin, C.: Helvet. med. acta **13**:595, 1946.
10. Hemmeler, cited by Undritz and Röthlin.[9]
11. Sabin, F. R.: Bull. Johns Hopkins Hosp. **34**:277, 1923.

little groups of checkered pattern (*Felderung* of Seeliger [12]) provides the centers of disintegration of the mature cell. I have directed attention to the phenomenon of dissociation in which the nucleus of the megakaryocyte develops to normal maturity while the cytoplasm remains in the basophilic stage without a trace of a functional area. In no such instance could either a segregation of single thrombocyte-like bodies or a disintegration of the cytoplasm be observed. Therefore, it must be concluded that there is no justification for assuming either a juvenile nongranulated stage of the thrombocyte in physiologic conditions or a pathologic inhibition of that alleged development. The examples referred to by Juergens and Graupner [2c] do not, in my opinion, substantiate their system, since their juvenile stages cannot be traced back to megakaryocytes. Their illustrations must be interpreted as portraying artefacts according to the observations described in this paper. The source of these structures interpreted as "juvenile" or "pathologic" basophilic thrombocytes is to be looked for in other cells than the megakaryocyte. This was particularly emphasized by Castranuovo,[13] who traced these bodies back to any type of blood cells and called them "pseudo-platelets."

SUMMARY

In its early developmental stage the megakaryocyte is enveloped by a layer of a viscous material outside the cell contour. This envelope disappears with advancing maturation of the cell.

In the preparation of smears this thin layer is spread, lacerated and sheared off. Structures simulating ameboid pseudopodia result from the application of those forces. Such an origin of the pseudopodia is substantiated by the features of the excrescences. Their mechanical separation from the cell simulates the formation of thrombocytes. These pseudothrombocytes lack a chromomere. The hypothesis that a chromomere is formed in the protrusions is based on a wrong interpretation of appearances produced by the apposition and superposition of true thrombocytes.

This demonstration rules out any thrombocytopoietic activity of the basophilic early stages of the megakaryocyte. The uniformity of the production of thrombocytes, which is thus restricted to the granulated mature stages, is therefore restored.

Most of the structures described as "juvenile" or "pathologic" thrombocytes are pseudothrombocytes, which can be traced back to various basophilic, ungranulated stages of various types of blood cells.

12. Seeliger, S.: Folia haemat. **29**:23, 1923.
13. Castranuovo, G.: Haematologica **1**:474, 1920.

ADRENAL PHEOCHROMOCYTOMA

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FIFTY years ago (1899) pheochromocytoma of the medulla of the adrenal gland was recognized as a distinct entity at a postmortem examination by Robert. Nearly thirty more years passed before the tumor was diagnosed during life, and Charles Mayo's report of successful surgical removal quickly followed in 1927. Soon after this, in 1929, the epinephrine content of a tumor was estimated by Rabin. Since that time, further reports and reviews have been published which have added to knowledge of adrenal chromaffinoma or pheochromocytoma.

A case of pheochromocytoma of the medulla of the adrenal gland will now be described on account of the unusual size of the tumor and because studies have been made on its epinephrine content.

REPORT OF A CASE

Miss M., aged 58, was in good health until a year before admission, when tachycardia and nocturnal dyspnea developed. Her physician regarded these symptoms as manifestations of cardiac failure, the cause of which he could not determine. The heart was not enlarged, no murmurs could be heard, and the blood pressure was not abnormal. For a week before admission the patient frequently wanted to pass urine but was unable to do so. On the day of admission her breathing became much more labored. At the time of examination she was semicomatose, and her skin felt cold and was of a mottled purple color; her temperature was 95.6 F. The pulse rate was 118; its rhythm was regular. The blood pressure reading was 105 systolic and 70 diastolic. The apex beat was in normal position, and no abnormal cardiac sounds were heard. The veins of the neck were considerably distended. The edge of the liver was just palpable. No subcutaneous edema could be detected. The respirations were increased in rate and depth. There was some diminution of the percussion note at the bases of the lungs. Crepitations, rales and rhonchi could be heard over the whole of the chest. In the left hypochondrium a tumor was felt, which was not tender. The pupils were dilated and of regular outline. No other physical signs were detected. Glycosuria was not demonstrated. The patient failed to respond to resuscitative measures and died twelve hours after admission.

Postmortem Examination.—This was performed nine hours after death in midwinter. On the left side of the abdomen a spheroid tumor was present, measuring 17 by 15 by 6 cm. and weighing just over 1,200 Gm. (fig. 1). It lay above the

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left kidney, which was depressed down to the rim of the pelvis. The spleen was stretched across the left side of the upper portion of the tumor, and the body and tail of the pancreas across its medial aspect. The tumor appeared to be covered completely by fibrous connective tissue. It felt partly cystic, partly firm. On section it was found to consist of purplish fleshy tissue, mottled gray in places, and was separated into lobules by strands of fibrous tissue. In places, chiefly centrally, orange masses occurred, which contained cystic spaces. The cut surface bulged considerably. No trace of normal adrenal cortex could be seen. A summary of the other postmortem observations follows. Small bilateral pleural effusions were present. Both lungs showed apical emphysema, congestion and at the bases subpleural collapse. The heart weighed 340 Gm. There was slight subendocardial fibrosis of the left ventricle. The left anterior descending coronary artery showed atheromatous narrowing and was calcified in places. The aorta was moderately atheromatous. The liver appeared a little congested. The thin-

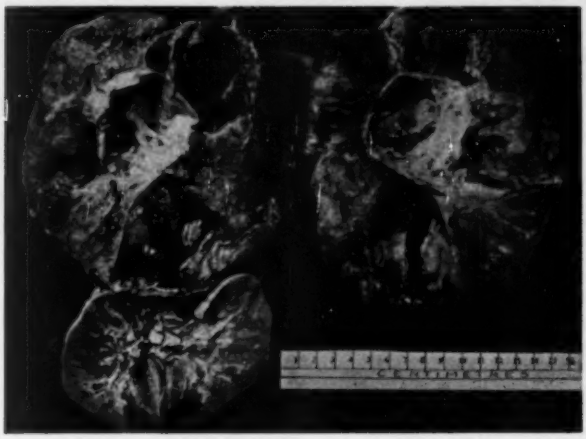

Fig. 1.—Tumor of the medulla of the adrenal gland and the left kidney incised in the long axis.

walled gallbladder was distended because of obstruction of the cystic duct by a faceted gallstone. There were many similar small gallstones in its cavity. There was slight congestion of the spleen and the kidneys. The right adrenal gland appeared natural. Death was considered to be due to acute cardiac failure arising as a result of pheochromocytoma of the medulla of the left adrenal gland.

Histologic Examination.—Hematoxylin-eosin sections of formaldehyde-fixed material were first examined. The cells of the tumor were of variable size. They were polygonal in shape and in places formed syncytial masses. The cytoplasm was for the most part abundant, lightly to strongly eosinophilic and finely granular. The nuclei showed considerable variation in size and shape, some reaching giant proportions (fig. 2*A*). A few of the cells contained more than one nucleus, one cell being observed with six. The staining of the nuclei was a little uneven. The nuclear membranes were distinct and the chromatin tended to be coarsely aggregated. In a proportion of cells nucleoli could be made out. Mitotic figures were

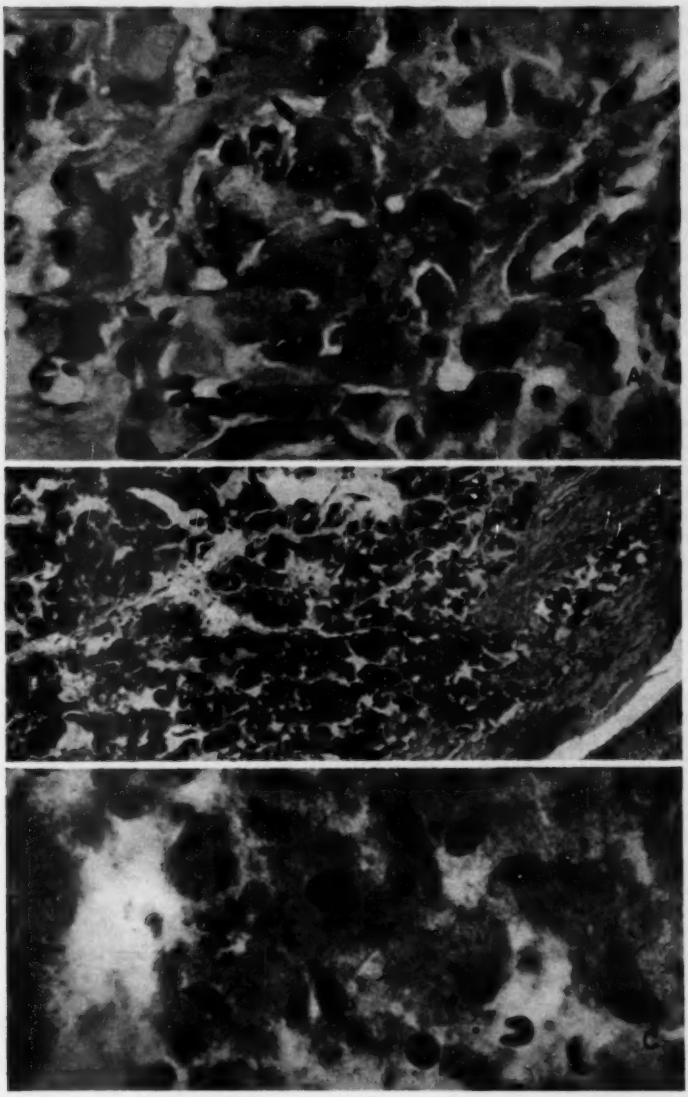

Fig. 2.—*A*, high power photomicrograph illustrating the pleomorphism of cells in some areas. Hematoxylin-eosin; × 470.

B, section of the tumor's edge, showing tumor cells infiltrating the fibrous capsule. Hematoxylin-eosin; × 100.

C, intracytoplasmic granules of epinephrine. Cramer's method; × 1,300.

rare. Basophil granules were seen in a few of the cells, being distributed usually at the periphery of the cytoplasm. The tumor tissue was intersected by fibrous trabeculae containing large blood vessels; otherwise the stroma was scanty, consisting of capillary blood vessels and patchily distributed thin strands of fibrous connective tissue. The tumor was surrounded by a fibrous capsule of variable thickness, which contained blood vessels, mostly of capillary size. Projections of tumor tissue extended into this capsule, and in places aggregates of cells lay apparently separated from the main mass (fig. 2*B*). Within the tumor there were areas which showed varying amounts of degeneration. In some of these the cells had been replaced by a hyalinized loose fibrous connective tissue; in others, by an edematous and sometimes hemorrhagic matrix. Associated with one of the latter areas was a venule which showed a recently formed thrombus. A frozen section stained with scarlet red indicated that some of the tumor cells in this area had undergne fatty degeneration. Parts of the tumor showed a strong histologic resemblance to normal medullary tissue.

Stains for mucin, glycogen and alkaline phosphatase gave negative results. No nerve tissue was recognized in the substance of the tumor.

Chromaffin Reaction.—When a piece of tumor tissue was immersed in a chromium salt fixative,[1] it became a rich brown within a few hours. A section of tissue fixed thus was stained after the method of Wiesel. Difficulty was found in producing any staining with the toluidine blue solution. Under oil immersion greenish brown granules of variable size could be seen lightly distributed throughout the sections. They lay usually in a paranuclear position. Another section fixed in the same way was stained by Schmorl's method for chromaffin cells. The cytoplasm of all the cells stained bluish green. In addition, globules of the same order as in Wiesel's preparation were present in the cytoplasm. These were of a more intense blue-green color.

It was further observed that the mass of tumor which was preserved in 4 per cent formaldehyde solution but otherwise exposed to the atmosphere changed from its initial purplish color to a deep rich brown in the course of two days. This became more intense on further standing, and the fixative also became stained this color. The tumor was later transferred to Pick III solution, and successive changes of this during the next four months were likewise stained brown, though with diminishing intensity.

THE DEMONSTRATION OF EPINEPHRINE

Histologic Study.—The method of Cramer was applied to fat-free portions of the tumor tissue. The latter was fixed in osmic acid vapor, and some portions were afterward treated with turpentine. Black granules which were seen in the fixed and untreated specimens were unaltered by turpentine; in other words, these granules could be ascribed to epinephrine. They were usually in small clusters and visible only in a proportion of cells (fig. 2*C*). Their size and number approximated those of the chromaffin granules seen in the sections stained by the methods of Wiesel and Schmorl.

Chemical Studies of an Extract of the Tumor.—A portion of tumor weighing 44 Gm. was immersed in 50 cc. of 10 per cent trichloroacetic acid, cut up finely and allowed to stand in the ice chest overnight. The final volume was made up

1. The formula of the chromium salt fixative used is as follows: 5 per cent potassium dichromate, 10 cc.; 4 per cent formaldehyde solution, 20 cc; distilled water, 20 cc.

to 120 cc. with distilled water, and the supernatant fluid was filtered. This filtrate was used for chemical studies and bioassay. It was found that the epinephrine in this filtrate was stable for at least six months.

Epinephrine was demonstrated in this extract by the following qualitative chemical tests. The extract gave a red color with the persulfate reagent of Ewing and a rose red color with iodine solution (Vulpian reaction), and it strongly reduced the uric acid reagent of Folin and Denis.

Three colorimetric methods were employed for the quantitative chemical estimation of the epinephrine present. A Coleman no. 11 spectrophotometer was used for this study. Pure crystalline epinephrine made up in hundredth-normal sulfuric acid was used for standard solutions.

1. Method of Folin, Cannon and Denis.[2]

Epinephrine may be estimated by its ability to reduce the uric acid reagent of Folin and Denis. Turbidity is prevented by adding urea. Details of the method used follow: To 1 cc. of solution containing epinephrine at a concentration between 1:50,000 and 1:200,000 were added 0.5 cc. of uric acid reagent, 1 cc. of 40 per cent urea and 4 cc. of saturated sodium carbonate solution. The blue color developed rapidly, was consistently reproduceable and began to fade in five minutes.

By this method the epinephrine content of the tissue was estimated to be 2.7 mg. per gram of tissue.

2. Persulfate method first described by Ewins[3] and modified by Barker, Eastland and Evers.[4]

Barker and associates described this as an almost specific chemical test for epinephrine in gland extracts, a red color being developed by the persulfate in the presence of epinephrine. They showed that maximum color developed in the presence of a chloride at a p_H of 5.5 and that although color was produced more slowly in gland extracts than in pure epinephrine solutions, maximum color was produced in half an hour at 22 C.

Following their conditions, we have obtained complete color production by pure epinephrine solutions only after one and a half hours, while the color produced by gland extracts continued to deepen over a period of eight hours or more. The ultimate color contained a definite brown component, which in time produced a brown precipitate. Barker and associates stated that the persulfate reaction is catalyzed by traces of copper salts but that the color so produced is uncontrollable and unstable.

By carefully following the color production in the presence of copper ions we have obtained highly reproduceable results with practically clear solutions. The technic was as follows.

To 4 cc. of solution containing epinephrine in a concentration between 1:10,000 and 1:100,000 were added 4 cc. of persulfate reagent and 2 drops of 1 per cent copper sulfate solution, and the p_H of the mixture was adjusted with 4 per cent sodium hydroxide to between 5.0 and 5.5, according to an external methyl red indicator. The persulfate reagent was prepared as follows. Potassium persulfate 0.2 Gm., sodium chloride 1.0 Gm., disodium phosphate (Na_2HPO_4) 0.1 Gm., monosodium phosphate ($NaH_2PO_4 \cdot H_2O$) 1.0 Gm., and water to 100 cc. Readings of absorption at a wavelength of 500 millimicrons were made at frequent intervals until the maximum color had begun to fade.

2. Folin, O.; Cannon, W. B., and Denis, W.: J. Biol. Chem. **13**:477, 1913.
3. Ewins, A. J.: J. Physiol. **40**:317, 1910.
4. Barker, J. H.; Eastland, C. J., and Evers, N.: Biochem. J. **26**:2129, 1932.

It was found that the hydrogen ion concentration markedly influenced the rate of color production and fading but that the maximum intensity of color produced was not affected. Also, the color produced was proportional to the concentration of epinephrine present and obeyed Beer's Law. It seemed, therefore, that this modified procedure could be used to estimate reliably the amount of epinephrine in the gland extract.

By this method the epinephrine content of the tissue was found to be 3.8 mg. per gram of tissue.

3. Method of Shaw.[5]

Shaw described a method whereby reducing substances other than epinephrine were removed by aluminum hydroxide treatment. Glutathione is removed by aluminum hydroxide at p_H 4, while epinephrine remains in solution. Epinephrine is precipitated along with aluminum hydroxide at p_H 8 and is estimated by its reduction of a sensitive arsenomolybdic acid reagent. Shaw discovered that brief treatment with alkali enhances the reducing power of epinephrine and that this enhancement is specific for the side chain of epinephrine.

The aluminum hydroxide precipitations and alkali enhancement were carried out by us according to the aforenamed method. The concentrations of epinephrine used were adjusted to lie between 1:50,000 and 1:200,000, and the final blue solution was diluted to 50 cc. with distilled water.

Contrary to the findings of Bloor and Bullen,[6] no loss of epinephrine occurred during the aluminum hydroxide treatments when solutions of pure epinephrine were used. Also, our gland extract lost no reducing power on full treatment with aluminum hydroxide. Although the degree of enhancement was not always reproduceable, the special alkali treatment caused similar increases in color in both our gland extract and solutions of pure epinephrine.

The epinephrine content of the tumor was found by this method to be 2.7 mg. per gram of tissue.

A pair of normal adrenal glands analyzed by these three methods was found to contain the following amounts of epinephrine:

(1) Folin, Cannon and Denis.....4.0 mg.
(2) Barker, Eastland and Evers.....2.9 mg.
(3) Shaw .....2.4 mg.

Since the method of Folin, Cannon and Denis is affected by the other reducing substances normally present in the adrenal gland, it would be expected to give the highest values.

The discrepancies between the results obtained by the persulfate method and that of Shaw are similar for both tumor and normal gland. Since perfectly clear solutions were not obtained in the persulfate method, it is likely that the results obtained thereby were erroneously high. Shaw's method must therefore be considered as being the most suitable for the chemical estimation of the epinephrine of tissue extracts.

Since solutions of pure epinephrine and tumor extract behaved in identical manners in the procedure of Shaw, and as the values obtained for epinephrine content of the tumor by the three methods were approximately the same, it seems reasonable to conclude that 2.7 mg. per gram of tumor represents the actual concentration of epinephrine and that no epinephrine-like compounds are present.

5. Shaw, F. H.: Biochem. J. **32**:19, 1938.
6. Bloor, W. R., and Bullen, S. S.: J. Biol. Chem. **138**:727, 1941.

If it is assumed that epinephrine is distributed evenly throughout the tumor, the total amount of this substance is calculated to be 3.2 Gm.

Bioassays.—An aliquot of the tumor filtrate was sent to Mr. F. N. Fastier, M.Sc., of the department of medicine of the University of Otago, who undertook to carry out bioassays by two methods. One of these made use of a rat hindquarter preparation.[7] Fluid was perfused through this, and the change in perfusion pressure brought about by the addition of tumor extract was compared with that produced by a standard epinephrine solution. This method gave the strength of the extract as 1 in 5,000. The other technic involved the inhibition of contraction of smooth muscle of rabbit intestine by epinephrine. The strength of the extract by this method was 1 in 3,500. These results of bioassays are equivalent to concentrations of 0.6 mg. and 0.9 mg. of epinephrine per gram of tumor tissue.

Mr. Fastier considered that the results might have been affected in some way by the solvent used.

COMMENT

The number of cases of pheochromocytoma which have been reported is expanding rapidly. In review of the world's literature Calkins and Howard[8] obtained a total of 176 such tumors. As a result of the clarification of the clinical picture, well summarized by Cahill,[9] an increasing number of cases are being diagnosed in life. The most striking modes of presentation are the result of paroxysmal hypertension. On occasions, however, the hypertension may be persistent,[10] and it has been reported malignant (i. e., rapidly progressive).[11] The diabetic features may be to the fore[12] and sometimes symptoms and signs suggest a diagnosis of thyrotoxicosis.[13]

However, as our case illustrates, there has been no good correlation between the severity of symptoms and the epinephrine content or the size of the tumor. Apart from the case of Borch-Johnson (cited by Biskind, Meyer and Beadner[14]) ours appears to be the heaviest tumor yet reported, being 200 Gm. in excess of that of Belt and Powell.[15] Except for the large quantity reported by the latter authors, the total epinephrine content of our tumor, 3.2 Gm., is apparently the greatest amount that has been found, and yet the disturbance

7. Fastier, F. N., and Smirk, F. H.: J. Pharmacol. & Exper. Therap. **89**:256, 1947.

8. Calkins, E., and Howard, J. E.: J. Clin. Endocrinol. **7**:475, 1947.

9. Cahill, G. F.: J. A. M. A. **138**:417, 1948.

10. Thorn, G. W.; Hindle, J. A., and Sandmeyer, J. A.: Ann. Int. Med. **21**: 122, 1944. Green, D. M.: J. A. M. A. **131**:1260, 1946.

11. Gutmann, D.: Brit. M. J. **1**:563, 1947.

12. (*a*) Duncan, L. E.; Semans, J. H., and Howard, J. E.: Ann. Int. Med. **20**:815, 1944. (*b*) Goldner, M. G.: J. Clin Endocrinol. **7**:716, 1947.

13. Cabot Case 32511, New England J. Med. **235**:906, 1946.

14. Biskind, G. R.; Meyer, M. A., and Beadner, S. A.: J. Clin. Endocrinol. **1**:113, 1941.

15. Belt, A. E., and Powell, T. O.: Surg., Gynec. & Obst. **59**:9, 1934.

to the patient was minimal. The patient did not complain of symptoms until a year before death; when symptoms developed, they consisted mainly of palpitations and shortness of breath, and until the terminal stage they were considerably less distressing than those which have occurred in other reported cases. The structure of the tumor suggests that growth had been taking place over a number of years. It is possible that a tolerance of the increasing amounts of epinephrine discharged into the blood stream gradually developed. Insensitivity to epinephrine, disappearing after removal of the tumor, has in fact been reported.[16]

The macroscopic appearance of the tumor is similar to that of tumors of the same type described by Belt and Powell[15] and other authors. Histologically, too, the picture is similar to the descriptions given in other reported cases, e. g., that of Edward.[17] As some tumors show a more regular cytologic pattern than others, they might be expected to secrete more epinephrine per unit mass than the latter. The cellular pleomorphism seen in this case (fig. *2 A*) is not considered by Edward[17] to be an index of cancerous change, but this feature together with evidence of capsular infiltration (fig. *2 B*) must be taken as indicating that our tumor was growing somewhat atypically. The amount of epinephrine per gram, 2.7 mg., was, in fact, considerably less than the amounts reported in a number of other cases.[18]

Basophilic cytoplasmic granulations such as those observed in some cells of our tumor have been noted by Mayock and Rose[16] and Duncan and co-workers.[12a] The latter commented on their resemblance to the Nissl granules of ganglion cells. No nerve elements could be recognized in our sections. In contrast to the findings of Muir,[19] no glycogen could be detected in the tumor cells. Fat globules could be demonstrated only in areas of degeneration. We were more fortunate than Edward[17] in being able to demonstrate epinephrine in the cytoplasm of some of the cells by the method of Cramer.

The chromaffin reaction was strongly positive. The term has been used in the literature to describe two features, the brown staining of cells treated with chromium salts and the intracytoplasmic brown granules demonstrated after chromium salt fixation. It has not been made clear whether the diffuse and the granular staining are in any way related. The former has been ascribed to the browning of intra-

16. Mayock, R. L., and Rose, E.: Am. J. M. Sc. **213**:324, 1947.

17. Edward, D. G.: J. Path. & Bact. **45**:391, 1937.

18. (*a*) Wells, A. H., and Boman, P. G.: J. A. M. A. **109**:1176, 1937. (*b*) Spalding, J. M. K.: Brit. M. J. **1**:565, 1947. Belt and Powell.[15]

19. Muir, R.: Textbook of Pathology, ed. 5, London, Edward Arnold & Co., 1941, p. 965.

cellular epinephrine,[20] whereas other authors[18a] have considered that the granules appear when the chromium salts have been reduced by the epinephrine to an insoluble peroxide of chromium.

Our tumor became stained a deep brown quickly on addition of potassium dichromate, but it also showed the same color change, more slowly, after being exposed to air. The numbers of chromaffin granules which we were able to show by the methods of Wiesel and Schmorl were of the same order as that of the epinephrine globules shown by the method of Cramer.

It seems likely, therefore, that the potassium dichromate merely greatly accelerates the oxidizing of epinephrine to a brown compound which will appear in any case at a slower rate in the atmosphere. That chromium enters into the composition of the granules is suggested by the following experiment: When equal parts of 0.1 per cent epinephrine solution and potassium dichromate fixative were mixed, the solution first became red and then dark brown, and in a few minutes a brown precipitate formed. After the latter had stood for a few days, it was centrifuged and washed with distilled water. On analysis it was found to contain both organic matter and chromium.

SUMMARY

A case in which pheochromocytoma of the medulla of the adrenal gland was diagnosed at postmortem examination is described. The tumor was large, weighing 1,200 Gm. Chemical and biologic assays of epinephrine were made on extracts of this tumor. The relative merits of the methods for the chemical estimation of epinephrine were investigated, and it was found that the technic of Shaw was the most suitable for this purpose. By it the total amount of epinephrine in the tumor was calculated to be 3.2 Gm. The chromaffin reaction is discussed.

20. Carleton, H. M., and Leach, E. H.: Histological Technique, ed. 2, London, Oxford University Press, 1947, p. 289.

GENIC FACTORS IN VISCERAL ASYMMETRY AND IN THE DEVELOPMENT AND PATHOLOGIC CHANGES OF LUNGS, HEART AND ABDOMINAL ORGANS

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SITUS inversus viscerum is one of the most fascinating of the anomalies of man. To judge from the number of papers on the subject, interest in it is equaled only by that in the transposition of the great vessels, the puzzling anomaly correlated to situs inversus. The intention of the present paper is to give further insight into these problems on the basis of a study of the genetics and the developmental genetics of situs inversus.

Situs inversus is no simple alternative to the normal situs. It is the most complicated among anomalies, concerning not only the situs, but all details of structure. There are a great many studies of the anatomy of situs inversus, culminating in the papers of Pernkopf (1937).

The problem of situs inversus is a part of the problem of symmetry and has called on the interest of medical men, anatomists and other scientists in the different fields of biology. The literature on situs inversus and related topics is immense, therefore, and in this brief survey I can deal only with a few previous studies that are essential to an appreciation of the problems of the present paper.

Spemann and his pupils found the anomaly in about one half of the twins which were derived from the right halves of eggs divided mechanically in different stages of development. They also produced inversion of the heart by rotating pieces of the roof of the primitive gut. Komai (1938) found in salmon a high correlation between situs inversus, small size and malformations which occurred in single individuals as well as in separate and in conjoined twins, presumably due to the environmental factors in the fish hatcheries.

Situs inversus was first found as a genetic variation in experimental animals in 1948, when Tihen, Charles and Sippel published their observation of situs inversus as caused by a recessive gene in mice. As in man so in these animals there were several associated anomalies, particularly hydrocephalus. They found 29 affected individuals and 227 normal individuals among the offspring of heterozygotes, about one

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half of the expected number. They suggested that this may be due to prenatal selection or to some variation of the phenotype. Nothing is known as yet about the hereditary and developmental relationship between situs inversus and the associated anomalies in mice.

Several cases are on record showing that the condition may be hereditary in man. Mattison observed the anomaly in mother and daughter, while Pernkopf (1937) found complete situs inversus in a father and partial situs inversus in his child. Mattison examined 448 members of the families of 4 patients without finding it in any and Gutzeit and Lehmann had the same experience in an extensive examination of the families of 3 patients.

As far as I know, 29 families are on record with more than 1 sibling affected—7 with 3 siblings affected, 22 with 2 affected. (Cockayne, 1938; Gänsslen, Lamprecht and Werner). Two of the families in which 3 were affected and 2 of those in which 2 were affected were reported from Norway, by Frölich, Torgersen (2 cases, 1946) and Natvig.

The only investigation based on a large number of cases is Cockayne's (1938). His paper represents the most important approach to the genetics of situs inversus. Cockayne found 58 affected and 90 normal persons in 52 reliable sibships. This gives, according to Cockayne's calculation, a ratio of affected to normal of 1 : 1.7, which represents, as far as I can see, the ratio of the index cases to the normal siblings. In a review of the literature he found a ratio of affected to normal of 1 : 2.9 in 119 sibships with 137 affected and 402 normal siblings. Cockayne concluded, therefore, that the anomaly is determined by a single recessive gene. He interpreted the associated anomalies as expressions of a manifold effect of this gene. Gutzeit and Lehmann admitted the possibility that there was dominant inheritance in some cases.

As far as I know, situs inversus has been observed 12 times in monozygotic twins, 6 times concordant and 6 times discordant (Cockayne, 1938; Gänsslen, Lamprecht and Werner; Kean; Helweg-Larsen). According to Cockayne (1938), discordant situs inversus has been observed once in dizygotic twins.

Mattison made an extensive investigation of the occurrence of twins in the families of persons with situs inversus and found an insignificantly higher percentage than in the population. There are on record a few cases indicating a relationship between twinning and situs inversus. Doolittle reported a case in which a probable isolated dextrocardia occurred in a pair of dizygotic twins, the father of whom was a dizygotic twin with dextrocardia. There were many twins in the family on both the paternal and the maternal side. Helweg-Larsen reported that there were several twins in the family in his case.

Joyce asked in the *British Medical Journal* whether it depended on chance that he had observed 2 cases of situs inversus in which there were surprisingly many twins in the families, both parents being twins in 1 case. In Reinhardt's case (1912) the mother of the twins with situs inversus was a twin and she and their father were first cousins. Most remarkable are the families in which parents who are twins have twins showing situs inversus.

Cockayne (1938) expressed the opinion that the frequent concordance noted in one egg twins is evidence of heredity. In 1939, in view of the observation of a discordant monozygotic pair of twins, he admitted the possibility that a mirror image effect occurs in cases of late division. This possibility is confirmed by the fact that all the cases of isolated dextrocardia on record are discordant; 3 of the patients were separate twins and 2 were conjoined twins. In the discordant cases of Dubreuil-Chambardel, Cockayne (1939) and Helweg-Larsen there is some dissimilarity of the twins, the first pair showing mirror image harelip and the second mirror image asymmetries indicating a late division. These twins may have narrowly escaped being conjoined twins.

Cockayne found first cousin marriages in 11 per cent of 53 cases, and he emphasized that even 6 per cent would be a convincing proof that the condition is determined by a single recessive gene. Cockayne originally suggested the idea of recessive inheritance in the discussion concerning the familial case of Feldman, in which the parents were first cousins.

A supposition of a particular mechanism of inheritance increases the risk of selection. I have found first cousin marriages in only 2 of the 29 familial cases of situs inversus in the literature (Ochsenius and Feldman). There may be some more but I have not had an opportunity to study all the original papers.

Situs inversus may be associated with bronchiectasis and nasal polyps (Kartagener; Olsen; Torgersen, 1947). There is good evidence that bronchiectasis may be inherited. The most reasonable interpretation of the data is that the genes causing bronchiectasis behave in the same way as genes showing incomplete dominance. The syndrome has been observed 3 times in father and child, 10 times in siblings, 4 times in both of a pair of monozygotic twins and 2 times in one of a pair of dizygotic twins (Kartagener; Diehl). Familial occurrence of situs inversus and bronchiectasis has been observed by Cockayne (1938). In a sibship of 5 bronchiectasis was found in one sib and situs inversus in another. Jerman (cited by Diehl) observed situs inversus in a mother and bronchiectasis and chronic ethmoiditis in her son. Kartagener reported situs inversus occurring in one brother and situs inversus and bronchiectasis in another. These observations are

more indicative of genes causing bronchiectasis as a primary effect and situs inversus as an expression of a pleiotropic effect than of genes causing situs inversus as a primary effect and bronchiectasis as a secondary effect.

Adams and Churchill had examined the patients of the Massachusetts General Hospital since the year 1886, numbering in all 232,112. Bronchiectasis occurred in 0.36 per cent; situs inversus, in 0.01 per cent. Bronchiectasis occurred in 21.7 per cent of the persons showing situs inversus; situs inversus occurred in 0.7 per cent of the persons showing bronchiectasis.

The previous observations of the frequency of bronchiectasis in persons with situs inversus are set forth in table 1.

The agreement among the authors as to the frequency of situs inversus in cases of bronchiectasis and of bronchiectasis in cases of situs

TABLE 1.—*Recorded Instances of Situs Inversus Complicated with Bronchiectasis*

		Patients	Per Cent
Horlacher (cited by Diehl):	Situs inversus	30	
	Bronchiectasis with situs inversus	7	23.3
Olsen:	Dextrocardia	85	
	Bronchiectasis with dextrocardia, 10 of whom had nasal polyps	14	
	Nasal polyps with dextrocardia without bronchiectasis	4	21.2
Torgersen (1947):	Situs inversus	129	
	Bronchiectasis with situs inversus, 5 of whom had nasal polyps	10	
	Nasal polyps with situs inversus without bronchiectasis	6	
	Severe bronchitis or frequent pneumonia with situs inversus	13	22.4

inversus is evidence of the validity of the observations. Adams and Churchill discussed the difficulties in the making of the clinical diagnosis and in the separating of the congenital from the acquired defects. It is impossible to make a clinical differential diagnosis between severe chronic bronchitis, recurrent pneumonia and bronchiectasis. They stress the difficulty of establishing a correlation between these and such common diseases as sinusitis. One has also to consider that bronchiectasis is much more common than is generally assumed. Among the chronic diseases of the lungs it is second in importance only to tuberculosis.

Why do some people have bronchiectasis after one of the common nontuberculous diseases of the lungs and others not? This as a question of both heredity and environment. It is impossible to draw a sharp line between these two main factors in pathology as well as in genetics. Roberts mentions a family with many cases of sinusitis. He is inclined to mean that the familial sinusitis is due to particular genes and that it is, in consequence, etiologically different from the corresponding common disease.

In another preliminary paper (Torgersen, 1947) mention has been made of the rather frequent anomalies of the spine in persons with situs inversus.

Situs inversus is often complicated with malformation of the heart. This combination is so common that anatomic and etiologic studies of situs inversus and of congenital heart disease have to be based on this correlation. Spitzer, Aschoff and Pernkopf (1937) stressed the importance of this combination.

As may be seen, a study of situs inversus has to include the genetics and the pathology of common diseases. This makes the analysis of the material difficult. However, the rarity of situs inversus facilitates the establishment of a correlation between it and even rather common diseases. The motive of the present investigation has been not only an academic interest in situs inversus but also a practical interest from this particular point of view in the causation of these common diseases.

TABLE 2.—*Collection of Cases*

Mass roentgenography:	Number of persons	1,000,000
	Confirmed cases of situs inversus	104
	Frequency 0.01 per cent ± 0.0008	
Hospitals and physicians:	Cases of situs inversus, 10 of which are already included in the 104 cases from the mass roentgenography series	88
Sum total (104 + 88)		192
Expected number of cases of situs inversus in Norway		300
Not diagnosed, about		100
Family material:	Number of families	161
	Number of cases in these families	168

OWN INVESTIGATIONS

The data and an evaluation of the data are presented in this chapter. I have added some parallel observations from the literature, both from that concerned with human pathology and genetics and from that based on experimental embryology and genetics. In the last chapter I will discuss the bearing of the data on the problem of situs inversus and the development of the viscera and their predisposition to the diseases referred to in the foregoing pages.

My plan was to collect as many cases as possible from a small and geographically limited population. This decreases the risk of selection and also the risk of not detecting cases in sibships. As will be seen from table 2, about one half of the cases have been detected in a mass roentgenography series; the other half have been reported to me by physicians and from hospitals and institutions of public health. It may seem simple to diagnose situs inversus in mass roentgenography. There are some difficulties, however, which are familiar to people used to the technic. I have therefore included only cases in which there is no doubt about the diagnosis and cases in which the diagnosis has been confirmed by ordinary roentgen examination.

None of the numbers in the foregoing table can be regarded as quite exact. However, the errors are significant. Thus, situs inversus occurs in the population at a rate probably somewhat above 0.01 per cent. This corresponds to a total

number of 300 persons with situs inversus, about 66 per cent of whom have been detected. This makes it improbable that a significant number of cases, if any, are to be found in the sibships and among the parents of the persons with situs inversus. It is hardly possible to examine all the siblings personally, but there has been a great chance of detecting the anomaly in these families, which have been most interested in the present study, and most people in Norway have been examined by physicians, a great many also by roentgenologists. In two counties 85 per cent of all persons above 15 years of age have been examined by mass roentgenography. No familial case was detected in the examination of 139,596 persons, 17 of whom had situs inversus.

TABLE 3.—*Cases of Situs Inversus in Two Counties in Which Examinations Were Made*

Oestfold:	Number of persons examined	72,690	(84.17 per cent)
	Number with situs inversus	5	(0.007 per cent)
Opland:	Number of persons examined	66,906	(88.05 per cent)
	Number with situs inversus	12	(0.02 per cent)
Difference between the two counties....0.013 ± 0.008			

TABLE 4.—*Discrepancy Between Actual and Expected Incidence*

	Actual Number	Expected Number	d	d^2	d^2/e
Oestfold	5	8.5	3.5	14.44	1.6
Opland	12	8.1	3.9	15.21	1.9
					$X^2 = 3.5$
					$p = 0.06$

TABLE 5.—*Differences Between the Two Counties in Which Examinations Were Made*

	Oestfold	Opland
Density of population:		
Living in rural districts, per cent	81.5	54
Living in cities, per cent	30	7
Ratio first births to later births 1920/1930	28/72	25/75
Ratio first births to later births 1931/1941	42/58	35/65
Marriage frequency, per cent	6.42	5.07

The population in these two counties has been examined to such an extent that one gets some information about geographic variation, as shown in table 3.

An X^2 analysis considering the expected number of cases of situs inversus if the frequency had been the same in the two counties gives the result shown in table 4.

The difference probably depends on some other factor than chance, therefore. The two counties represent two extremes in the Norwegian population. The difference may be seen better from table 5 than from a description.

The urbanization of the population is more advanced in Østfold than in Opland, where the people to a great extent live in valleys separated by the high mountains. The proportion of first births is greater in Østfold and so is the marriage frequency. There are relatively many, but small, families, and the age of the mothers at birth of the children will be lower because the few children are born

in the first years of marriage. The frequency of the birth of twins as seen from table 6 shows a difference in the two counties. It is hardly possible to decide on the basis of the present knowledge whether this discrepancy between a district with many isolated groups and a district with far advanced urbanization is due to inbreeding and genic factors or to the breeding habits of the population, expressing itself in the age of the mothers at birth, or to unknown factors.

In these counties 97 per cent of the births have been registered. It is improbable that there are so many twins in the remaining 3 per cent that this will influence the frequency in the statistics.

The correspondence between the occurrence of situs inversus and that of twin births may be evidence of a causative relationship. However, both may be due to independent factors tending to be relatively common in isolated groups.

The size of the population is 3,000,000. Supposing the frequency of twin births to be about 1.4 per cent, one should expect about 60,000 twins in the country, considering the high infant mortality, about 45,000 dizygotic and 15,000 monozygotic twins. Supposing a frequency of situs inversus of 0.1 per cent, one would expect 1.5 monozygotic twins with situs inversus and 4.5 dizygotic twins with the anomaly. I have actually found 2 possibly monozygotic twins, as well as 2 dizygotic twins, with situs inversus—the latter 2, surprisingly, both

TABLE 6.—*Occurrence of Twins in the Two Counties Examined*

In Oestfold:	1920-1930	1.5 per cent
	1931-1941	1.1 per cent
In Opland:	1920-1930	1.6 per cent
	1931-1941	1.5 per cent

Difference of Opland and Oestfold averages: $1.55 - 1.3 = 0.25 \pm 0.063$
Oestfold 1920-1930 and 1931-1941 percentages $= 0.4 \pm 0.3$

in the same pair. One of the monozygotic twins and one of the dizygotic twins with situs inversus was detected in the mass roentgenography series. Therefore, it is highly improbable that there should exist more twins with situs inversus in the country.

In one of the cases in which a probable monozygotic pair of twins was involved, the twin sister died at the age of 6 years. In the other case the twin sister died at the age of 1 month, and the statement of the mother that the twins showed great similarity does not mean much. There is no reasonable doubt that the third case concerned a pair of dizygotic twins (Torgersen, 1949).

Partial inversion of the particular type observed in 1 of these dizygotic twins occurred in 3 persons included in the present study, their number corresponding to a frequency of 0.0001 per cent. The probability that in this case the coincidence of twinning and situs inversus is due to chance is small. The chance that a sibling will show situs inversus, excluding the index cases, is about 1 per cent. The most reasonable assumption is that the inversion is partly due to some environmental factor, possibly causing a division in both of a dizygotic pair of twins. In other words, these twins may be survivors of quadruplets. The frequent occurrence of twin births in the family of the father is remarkable, considering the similar observation of Doolittle referred to in the introduction. The questions of the hereditary mechanism in twinning and the influence of the father are still unanswered.

Pernkopf (1937) was inclined to explain situs inversus as depending on some abnormality of the sperm, owing to his observation of visceral inversion

in father and child. It must be admitted that the observation of concordant situs inversus in dizygotic twins in a family with many twins on the paternal side may support his view.

There is no significant increase of the frequency of twin births in the siblings. I have reliable data about twins among the parents and grandparents in 30 families. In 2 cases one of the grandparents was a twin. This is not surprising. However, it is remarkable that in both these cases the patient with situs inversus was a twin, in one a monozygotic twin and in one a pair of dizygotic twins. In 1 case the mother was a twin and she and the father were first cousins. These observations in connection with the corresponding observations mentioned in the introduction are good evidence that there is a correlation between twinning and situs inversus in a few cases.

The data indicate that the frequency of monozygotic twins in cases of situs inversus does not deviate greatly from their frequency in the population. This

TABLE 7.—*Comparison of Ages of Mothers at Birth of Children*

The ages of mothers known in.....	164 cases of situs inversus
The ages of mothers known in.....	187 index cases
The ages of mothers known in.....	7 secondary cases
Mean age of mothers in cases (5,184 + 164).....	31.6
Mean age of mothers at birth generally.....	30.5
Difference	1.1 ± 0.46

TABLE 8.—*Actual and Expected Numbers of Mothers in Age Groups*

Age of Mothers	Percentage in Population	Actual Number		Expected Number		d	d^2	d^2/e
15-19	1.5	2	20	2.5	32.3	12.3	151.29	4.68
20-24	18.2	18		29.8				
25-29	28.4	46		46.6		0.6	0.36	0.01
30-34	23.8	42		39		3.0	9.0	0.23
35-39	17.8	56		29.2		6.8	46.24	1.58
40-44	8.9	15	20	14.6	16.9	3.1	9.61	0.57
45-	1.4	5		2.3				
		164		164				$X^2 = 7.07$
								$p = 0.10 - 0.20$

may be interpreted as evidence that in man monozygotic twinning plays no great part in the production of situs inversus. The numbers further indicate that the frequency of situs inversus in dizygotic twins is low, a supposition which is confirmed by the observations available in the literature.

In table 7 I have compared the age of mothers at the birth of children showing situs inversus with the age of mothers at the birth of children in the country generally. The numbers are small, because of the rarity of the anomaly, and difficult to evaluate, therefore.

In table 8 is seen the deviation between the actual and the expected numbers of mothers of children with situs inversus in the different age groups.

The difference between the age of mothers at birth of children with situs inversus and the age of mothers at birth of children generally is somewhat below 3 times the standard error. The X^2 analysis does not show any convincing evidence of a high age of the mothers. However, the consistent deviation in all age groups indicates a high age of the mothers.

The possibly high age of the mothers is due mainly to the 54 cases of situs inversus combined with bronchiectasis or congenital heart disease (table 9).

In table 9, also, the consistency of the deviation in all groups is conspicuous.

I have further analyzed the material to see if the order of birth has any influence. The child with situs inversus was the only child in 13 families. With these families excluded, it was the first-born in 29 families, the last-born in 34 families. Again, 440 children were born before and 334 children after the child with situs inversus. This difference is significant. As far as I can see there is no selective factor favoring the last orders of birth. According to Penrose (1934), there is reason to assume a selective factor favoring the first orders. In the families with more than two children in all, 20 per cent of the 270 first-born and last-born showed situs inversus, against 14 per cent of the 638 in the middle of the sibships. The difference is 6 ± 2.29. It is thus probable that the distribution of the children with situs inversus in the sibships is not due to chance alone.

TABLE 9.—*Numbers of Mothers in Different Age Groups Who Gave Birth to Children in Whom Situs Inversus Was Complicated with Bronchiectasis or with Malformation of the Heart*

Age of Mothers	Bronchiectasis	Hearing Malformation	Sum Total	Expected e	Deviation d	d^2	d^2/e	Familial Cases
15-19	0	0	0 } 4	11	7	49	4.45	2
20-24	3	1	4 }					
25-29	9	1	10	15	5	25	1.66	1
30-34	12	3	15	13	2	4	0.31	3
35-39	13	4	17	10	7	49	4.90	3
40-44	4	1	5 } 8	6	2	4	0.67	1
45-	3	0	3 }					
							$X^2 = 11.99$	
							$p = 0.02$	

The data, taken as a whole, are rather good evidence of an influence of the age of the mothers and a corresponding influence of the order of birth due to cases complicated with bronchiectasis and congenital heart disease.

According to Roberts, it is known that the age of mothers is high at the birth of children with congenital heart disease. Nothing is known about the age of mothers at the birth of children with bronchiectasis.

The lung-nose syndrome occurred in 4 of the 5 familial cases. In all these families except 1 the mothers were particularly old at the birth of the children with situs inversus. In the exception in which the mother was young there was evidence that the syndrome occurred in both parents. This may indicate that the influence of the age of the mothers in these cases concerns the manifestation of the genes.

The most reasonable conclusion is that the anomalies which are mostly influenced by the age of the mothers are the primary ones, the others, such as situs inversus, being expressions of the manifold effect of the genes. A parallel may be seen in cases of mongolism. The disturbance of the development of the heart is hardly the primary effect of the genic and nongenic factors, including the age of the mothers, but is secondary to the general disturbance of growth.

In 1 family the stature of the mother and 8 children was at the lower limit of normal. The last-born showed situs inversus and a congenital heart disease, probably the tetralogy of Fallot. The child next to him showed symptoms of idiocy. A sister showed retardation of the maturation of the bones and a short

middle phalanx of the fifth finger. The symptoms of mongolism are thus found in different members of the family. The father was of average stature, and the relative parts played by heredity and environment in this case are obscure. The living conditions of the family were poor, and the mother had given birth to 8 children by the time she was 32 years old. There is nothing remarkable about the physique of most of the persons with situs inversus. Constitutional anomalies have been recorded in some cases (Torgersen, 1948).

The average size of the 161 sibships was 5.9. I have compared the actual and the expected size of the sibships, supposing that the size of the sibships had been the same in cases of situs inversus as in the general population in the same years. There are significantly more large families in the cases of situs inversus. However, this is most probably due to the selection of large families in surveys in which only those families are recorded which contain affected persons (Penrose, 1934). The average size of the families is not surprising when one considers that a great many of the marriages took place about the year 1900 and that the average size of a Norwegian sibship in 1920, after twenty years of marriage, was 5.03.

In the total material 5 per cent of the children died before 1 year of age. The corresponding percentage in the population is 4.5 per cent. In the 40 families in which the lung-nose syndrome occurred 12 per cent of the 200 children died before 1 year of age. The difference between the percentage in this group and that in the total material is 7 ± 3. This may indicate a somewhat increased mortality in this group. As there is no sign of an increased mortality of infants in the other cases, it is reasonable to assume that the possibly increased mortality of infants of the group showing the lung-nose syndrome is due to a predisposition to diseases of the respiratory tract. A particular observation increases the probability of an increased mortality of infants in these cases. It is not included in the foregoing calculation. A woman with situs inversus was the last-born of 13 children, 10 of whom died before they were 1 year of age. One of her sisters had 13 children, 10 of whom died before they were 1 year of age. In another case the parents had a child with situs inversus, six lumbar vetebrae and spina bifida occulta. The mother gave birth a year later to a child with spina bifida aperta and had after that two abortions. In a third case a stillbirth occurred after eleven years of involuntary sterility following the birth of a child, the first-born, with situs inversus. In 2 more cases three stillbirths occurred besides the birth of a child with situs inversus. In one of the familial cases there was good reason to assume that sterility was present in the sibship itself, 5, possibly 6, siblings in a sibship of 8 being sterile. This family showed the lung-nose syndrome, the father and 4 children being affected. The parents were second cousins. An analysis of the pedigree does not indicate a linkage between genes causing these peculiarities.

The probably low frequency of situs inversus in dizygotic twins may according to Weinberg (1907), be a sign of prenatal selection. In some cases only one of a pair of dizygotic twins will have the lethal factor. The other will pass as not having been a twin, an event tending to decrease the number of dizygotic twins. The significance of these data is difficult to estimate. A prenatal mortality sufficient to explain the deficiency of affected persons when one assumes a single recessive gene presumes an improbably high mutation rate to compensate the corresponding loss of genes.

According to Roberts, there is no agreement among the statisticians as to what procedure is the best in order to avoid a selection of affected persons in a study of a character which is supposed to be due to a recessive gene. As

all the cases have been detected by an index case, I have excluded these cases as selected. The 2 familial cases which had been recorded previously should also have been excluded, thus making the number of secondary cases still lower. The number of index cases is 161, the numbers of affected and normal siblings are 7 and 777 respectively, giving a percentage of affected siblings of 0.9. The great deviation from the expected ratio in recessive inheritance is evident also from the fact that the index case is that of the only affected person in 156 families. It is also surprising that so few familial cases are on record. There is no reason to assume that this is due only to nondetection if one considers the extensive mass roentgenography carried on in many countries in recent years.

As the frequency in the population is about 0.01 per cent, the notion that the secondary cases are due to chance can be excluded. Murphy found that congenital malformations will occur twenty-five times as frequently as in the whole population in families in which the parents already have a malformed child. Situs inversus will occur one hundred times as frequently in families in which the parents have a child with situs inversus. This indicates a relatively strong influence of the genes in situs inversus. The number of index cases in the group of familial cases was 5, and the numbers of affected and normal siblings were 7 and 21 respectively. The fit to the mendelian ratio in these cases is probably not due to chance. The lung-nose syndrome occurred in 4 of these families—in both parents in 2 families, in one of the parents in 1 family. As mentioned, Kartagener reported the syndrome observed in a familial case. Lopez (1945) saw the syndrome in 3 siblings with situs inversus (cited by Torgersen, 1947).

In the mass roentgenography series the anomaly was once found in a second cousin. As it is no unreasonable assumption that the 161 sibships correspond to about 10,000 second cousins, 1 case may depend on chance. It cannot be excluded, however, that the anomaly depends on a recessive gene in this particular case. Cockayne (1938) reported 2 instances of situs inversus occurring in uncle and nephew. The same reasoning may be applied concerning these cases. However, it can hardly be excluded that the observations are due to selection, partly because the chance of detection is greater in a family in which the anomaly has already been detected in one member, partly because the risk of selection is great in a relatively small material collected from the vast population of England, Canada, South Africa and Australia.

The frequency of first cousin marriages in the total material was 3.1 per cent, of second cousin marriages 3.5 per cent and of marriages between remoter relatives 3.5 per cent. In families showing the lung-nose syndrome the frequency of first cousin marriages was 5 per cent ± 3.3, of second cousin marriages again 5 per cent. In the families not showing this syndrome the frequency of first cousin marriages was 2.5 per cent $\pm$ 1.5. Among the familial cases there was one in which the parents were second cousins. According to Dahlberg (1943), the frequency of first cousin marriages in cases of an anomaly caused by a single recessive gene and showing a frequency of 0.01 per cent is expected to be 3.5 per cent if the frequency of first cousin marriages in the population is 0.5 per cent. According to Dunn, Dahlberg has found the frequency of first cousin marriages in country districts in Sweden to be 0.45 per cent. Thus, the frequency of first cousin marriages is somewhat too low to fit with the supposition of a single recessive gene. If there is a multifactor mechanism, one should expect a relatively higher frequency of consanguineous marriages. The established frequency of consanguineous marriages in connection with the deviation from the mendelian ratio indicates the importance of homozygosity not of a particular recessive gene but of a complex of genes which may be conceived of as modifiers conditioning

the coming into existence of visceral inversion. These modifiers may be assumed to promote the development toward normal or toward abnormal asymmetry by influencing the reactive potency of the embryo. The manifestation of single genes or that of nongenic factors may be assumed to be conditioned by these modifiers and the environment. The deficiency of consanguineous marriages may be partly due to nonhereditary cases.

The most striking fact is the lack of first cousin marriages in the familial cases and the lack of secondary cases in the 5 families in which the parents are first cousins, no case of situs inversus being detected in 30 siblings. This is strong evidence against the supposition of a single recessive gene. According to Haldane, a deficiency of consanguineous marriages may be due to a recessive gene which has a lethal allelomorph. According to the foregoing discussion this assumption is improbable in this case. Cockayne (1938) expressed the opinion that the partial inversions, which rather often are lethal, probably depend on allelomorphs of the recessive gene that causes complete situs inversus. There is no evidence confirming this supposition. As will be dealt with in the last section, there is good evidence that the relationship between partial and complete inversion has other explanations.

The high frequency of the lung-nose syndrome in the familial cases and the low frequency of consanguineous marriages in the familial cases indicate that the association of situs inversus and this syndrome is not due to the fact that the chance of homozygosity concerning more than one pair of rare genes is greater in consanguineous marriages. Also this observation is in conformity with the supposition of the importance of homozygosity of modifiers, in this case of modifiers influencing the development of the lungs. In only 1 of the 12 cases from Opland County were the parents first cousins. This confirms the supposition that the high frequency of situs inversus in this county is only partly due to hereditary factors.

I did not find evidence of linkage of bronchiectasis and situs inversus (Torgersen, 1947). The data suggested that the syndrome was due to genic factors which in some cases cause situs inversus as an expression of a pleiotropic effect. This interpretation was based on the finding that the syndrome was more common in the siblings and parents in the cases in which the patient with situs inversus showed the symptoms than in the other cases. I also attached importance to the parallelism in the development of the frontal sinuses, which were small in cases of situs inversus associated with bronchiectasis and of average size in cases without the syndrome.

These observations and interpretations are confirmed in the greater material of the present study. The syndrome occurred probably in 40 families. In these families the frequency of situs inversus in the siblings was 3.7 per cent, against 0.16 per cent in the siblings in the families in which the syndrome did not occur. The difference, 3.54 ± 1.57, is indicative that situs inversus is more frequent in the families showing the syndrome. In 3 instances in which the families of both parents showed the syndrome, 3 of the 12 siblings showed situs inversus. The consistency of the deviations are evidence that the chance that a child is going to show situs inversus increases with the homozygosity of the genic factors in this syndrome.

In the cases in which the syndrome has occurred in the families of the parents, the frequency of the symptoms in the offspring is 56 per cent, against 26 per cent in the cases in which the syndrome has not occurred in the families of the parents. The difference 30 ± 10.6 may be regarded as significant, showing that

the frequency of the symptoms is higher in sibships deriving from a generation in which the syndrome has occurred.

The behavior of the genic factors of this syndrome is similar to the behavior of genes showing incomplete dominance. The relative frequencies of consanguineous marriages recorded in a foregoing paragraph confirm this supposition. Bronchography was performed in 5 cases of situs inversus. In 3 cases bilateral bronchiectasis was revealed. In each of these 3 cases one of the parents was affected. In 2 cases localized bronchiectasis was found. In these cases the parents were not affected. These few cases show parallelism to the genetic evidence.

As seen in the introduction, there is striking conformity between the observations of the present study and the observations of Olsen at the Mayo Clinic concerning the nasal polyps. Nasal polyps occurred in 22 per cent of the cases of situs inversus with bronchiectasis, against 5 per cent of the other cases of situs inversus. The difference 17 ± 10 indicates the same parallelism between the anomaly of the lungs and that of the frontal sinuses as was evident from the roentgen examination of the frontal sinuses. A frequency of nasal polyps of 5 per cent is hardly much above the frequency in the general population. However, the probability that this common condition belongs to the bronchiectasis and

TABLE 10.—*Variations of the Spinal Column in Cases of Situs Inversus*

Number of spines examined in cases of situs inversus	56	
6 lumbar vertebrae 10.7 per cent	23.2 per cent ± 5.8	
Lumbosacral vertebra 12.5 per cent		
Spines examined in parents and siblings	61	
6 lumbar vertebrae or lumbosacral vertebra	5	per cent ± 2.9
Spines examined in normal situs	500	
6 lumbar vertebrae 2.8 per cent	7.6 per cent ± 1.2	
Lumbosacral vertebra 4.8 per cent		

situs inversus is indicated by the alternating occurrence of bronchiectasis, nasal polyps and situs inversus in these families.

The observations are good evidence of genic factors in bronchiectasis and diseases of the frontal sinuses. The clinicians have to consider both genic and nongenic factors. It is a common opinion that sinusitis may be the cause of bronchiectasis. The data set forth in the foregoing paragraphs indicate that both conditions may have a common genic basis.

The variations of the spine in the cases of situs inversus are seen in table 10.

The difference between the percentages of persons with situs inversus showing six lumbar vertebrae or lumbosacral vertebra and persons with normal situs showing these anomalies is 15.6 ± 7. The corresponding difference with respect to persons showing situs inversus and their siblings and parents is 18.2 ± 8.7. The variability of the spine is probably increased in patients with situs inversus. This does not hold in regard to the siblings or parents. The data offer no evidence that genes affecting the spine are a cause of situs inversus. Anomalies of the spine were detected accidentally in 3 per cent of the total material. There were 3 cases of hemispondylus, 1 case of severe scoliosis and 1 case of coccygeal defect and imperforate anus.

Observations of a particular family indicate that the increased variability of the spine may have a genic basis. A woman with six lumbar vertebrae and a hypoplastic kidney showed situs inversus and spina bifida occulta. Two of her brothers had the same combination of anomalies of the spine and situs inversus. One of them had a lumbosacral vertebra, and this was also the case with a third brother showing normal situs and spina bifida occulta. The frequency of

this anomaly in cases of situs inversus was 18 per cent, against 9.6 per cent in cases of normal situs, a difference far from statistically significant. It is improbable that this occurrence of familial increased variability of the spine, spina bifida occulta, situs inversus and hypoplastic kidney in one of the siblings is coincidental. Observations of man (Torgersen, 1948) and of the mutations of the tails of mice show the correlation between anomalies of the urogenital organs and anomalies of the spine. Imperforate anus may be classified in this group of anomalies. These findings in the spine are of interest in view of the experimental inversion produced by rotating pieces of the roof of the primitive gut. As will be seen from the following statements, there is good reason to assume that the primitive vessels play an important part in inversion. Sawin has demonstrated a correlation between the variations of the spine and those of the aortic branches and their symmetry in the rabbit. Kühne (1931 and 1936) has shown the inheritance of the variations of the spine in man. Experimental evidence and observations of man suggest that variations in the development of the dorsal metameres play a fundamental role in the asymmetry of the viscera.

The morphology of the spine is of interest in the study of situs inversus not only because of the possibility that there is an inductive relationship between the dorsal region and the viscera but also because the heart and vessels take part in visceral asymmetry. The present material is selected as to the occurrence of heart defects. Such defects were noted in 10 cases in the total material and in 1 case in the mass roentgenography series. Cardiac defects occurred most probably twice in the 777 siblings. It is reasonable to refer briefly to the cases of heart defects even if the material is not representative, because these cases illustrate some of the characteristic defects. In the literature there are a great many observations demonstrating that the correlation of situs inversus, a variety of heart defects and malformations of the body is rather high. There was 1 person who showed defect of the ventricular septum, pulmonary stenosis and complete situs inversus. There were further a hemivertebra (third thoracic) and 13 ribs on the right side. Another had an abnormally symmetric liver, a gallbladder to the left of the ligamentum teres, multiple spleens, a defect of the ventricular septum and a patent foramen ovale. A third had a symmetric liver, coarctation of the aorta, a defective lower jaw, syndactyly and a defective first rib on the left. A fourth showed complete situs inversus, transposition of the large vessels, with the aorta "riding" over a defect of the ventricular septum, and stenosis of the isthmus aortae. A fifth showed symptoms and signs of the tetralogy of Fallot and isolated dextrocardia. A sixth also showed the tetralogy of Fallot and isolated inversion of the abdominal organs, these anomalies representing a mirror image of those of the fifth. As to the remaining ones, the diagnosis of the heart defect is doubtful. In all, 3 of the 158 persons with complete situs inversus without heart defect showed anomalies of the body and extremities, against 3 of the 10 persons in whom situs inversus was complicated with heart defect. Heart defect occurred in 5 of the 158 persons who had complete situs inversus, against 5 of the 10 who had partial inversion. These observations are in conformity with the great many previous observations showing that associated anomalies are more common in patients with heart defects than in patients with situs inversus and, further, with the observations showing that heart defects are more common in patients with partial inversion.

There is no convincing evidence of hereditary relationship of cardiac malformation and situs inversus in the present material, the incidence of congenital heart disease in the siblings being 0.26 per cent, certainly not more than in the population generally. In 1 instance a child with situs inversus and heart

defect was born after seven years of marriage and two previous abortions. A maternal great aunt, a son of another maternal great aunt and a sister of the paternal grandfather were said to have died of congenital heart disease at 11, 14 and 20 years of age. The observations in the present study are not evidence of genes causing localized anomalies of the body and situs inversus as an expression of a pleiotropic effect. In 1 case a brother and an uncle of the mother of the child with situs inversus had harelip and cleft palate. One of the siblings died from congenital heart defect when about 1 year of age. In another case a first cousin had harelip. In 1 case the mother showed a short fifth middle phalanx, as did a sister of the patient in the index case of the family just mentioned. This is not more than might be expected in cases of anomalies with a frequency of about 0.1 per cent and 1 per cent, respectively.

One of the persons with situs inversus had been operated on several times because of acute intestinal obstruction due to abnormal rotation of the mesentery. It is impossible to state how often anomalies of the abdominal cavity occur in clinical cases. Autopsies showed a relatively high correlation between such anomalies and situs inversus.

The stomach of the twin with inversion of the abdominal organs is of particular interest. The shape is rather peculiar, the pyloric part not ascending but taking a horizontal position. This observation indicates that the factors of asymmetry have an influence on the shape of the stomach.

Among the persons with situs inversus there were 78 females and 90 males. As the chance of detecting the anomaly has been somewhat greater in males, the sex ratio is probably about 1:1. Among the siblings there were 282 females and 367 males and 30 of unknown sex.

About 7 per cent of the persons with known handedness were left handed—probably not more than in the population generally.

COMMENT

There is no evidence that the hereditary mechanism of the asymmetries produced in Drosophila and snails has any bearing on the asymmetry of the viscera of man. There is no reason to assume that the recessive mutation observed in mice is the only cause of situs inversus in this animal. Probably the mutations which in a few years have been detected causing situs inversus in mice are even more numerous than the mutations and modifiers influencing the development of the tail. As will be seen from the following considerations there is good evidence that the gills and their derivatives, the heart and the lungs, are of particular importance in situs inversus. For this reason one has to be cautious in attaching weight to experimental analogies concerning causative factors. The relationship between the gills and the heart is different in man, fish and amphibia. The development of the lungs is fundamental in the asymmetry of the viscera and of the heart in man.

The observations indicate that there is a connection between situs inversus and twinning in a few cases, partly due to genic, partly to nongenic factors. There is good reason to assume that in some of the cases of discordant twinning this is due to a mirror image effect

caused by the late division. However, it is hardly possible to decide whether this mirror image effect is due to genic factors in bilateral differentiation. There is good reason to assume that conceptions as regulation, organizers, embryonic fields and inductive relationships are valid also in regard to man. However, it is difficult to decide at present to what degree such relationships depend on genic factors—a single gene or a few genes playing the part of the evocator, modifiers determining the reactive potency of the embryo.

The observations do not indicate a single factor mechanism as a general cause of situs inversus. It is reasonable to assume that the genic factors of bronchiectasis represent a very few of the genes responsible for the asymmetry of the viscera. Even if it is improbable that particular genes affect particular organs, the foregoing data are evidence that the development of the organs, as in the instance of the lungs, depends on particular genes, which at the same time influence the development of the asymmetry of the viscera.

In the third case in the casuistics cited in which a defective lower jaw was associated with an abnormality of symmetry and of the heart, the part played by the genes is obscure. The abnormality of the lower jaw and the asymmetry of the viscera in that case are not coincidental. Gruber found situs inversus in 6 of 82 cases of defective development of the lower jaw. The cases often showed multiple malformations of the heart and the extremities. Mohr reported a similar case in which the parents were first cousins.

Pernkopf (1926), from a morphologic point of view, concluded that the development of the viscera and the asymmetry of the organs were due to innate growth tendencies of the particular organs. That conclusion is supported by this study of the genetics of visceral inversion. Genes affecting the lungs may cause situs inversus in the same way as those affecting the lower jaw and the spine. The complex of genes affecting the organs and their asymmetry may be conceived of as modifiers. The present study is an approach to a breaking up of this complex of genes which are responsible for the asymmetry of the viscera.

Spitzer came to the conclusion that situs inversus depends on the structure of the cytoplasm, an opinion which is rather common among biologists (W. Ludwig; Harrison). The observations in man are in conformity with the assumption that the reactive potency of the embryo is one main factor in asymmetry reversal. The single gene or the genes which upset the balance play the role of the evocator. The observations indicate that the reactive potency depends on genic factors. As to the role of cytoplasm, they do not permit any conclusions.

The similarity between the development of sex and symmetry has been dealt with by W. Ludwig. He discussed the effect of the genic influence with respect to symmetry in terms similar to those used

by Goldschmidt in theorizing about the development of sex, the factors Right and Left taking the place of F and M.

The not infrequent combination of localized anomalies such as harelip and visceral inversion or transposition is evidence that in man there is a correlation similar to that found by Komai in salmon. There is no evidence that localized anomalies caused by a relatively simple hereditary mechanism are combined with these malformations due to a pleiotropic effect of these genes. They may be considered as phenocopies, caused mainly by nongenic factors. The identical results of genic and nongenic factors demonstrate the fundamental role of the competence of the embryo. Such combinations are seen also in double monsters, and it is probable that the case of discordant situs inversus with mirror image harelip belongs to this group.

The data indicate that the factors modifying toward normal asymmetry are decisive in the development not only of the topography of the organs but also of the structure and of normal and pathologic function. This assumption was reasonable concerning the lungs and, as will be seen from the following considerations, a similar chain of causes is probable in regard to the development of the heart and of the abdominal organs.

It is of interest that the observations lead to the same conclusions which Landauer (1948) reported regarding the asymmetric expression of the genes in polydactyly of fowl. This study shows that the asymmetries of the body may have a genic basis. Sinistral expression of these genes gives response to selective breeding in the same way as bronchiectasis and situs inversus in cases of hereditary bronchiectasis. Landauer assumed that the asymmetric expression depends on an influence exerted on growth differentials by multiple genic factors. Lateral growth asymmetries may be supposed to be influenced by modifiers. This concept of modifying genes reveals the integrative interaction between the mutant gene and specific developmental potentialities of the embryonic field in which the gene takes effect. As will be seen, this reasoning and the reasoning concerning the observations in the present paper are in conformity.

The sporadic occurrence of situs inversus in connection with lack of signs of phenotypical expressions explaining a deficiency of ratio compared with the expected mendelian ratio, the possibly high age of the mothers and the concordance in a pair of dizygotic twins may be regarded as evidence that in not a few cases situs inversus is due to nongenic factors. The correlation with the malformations of the heart in which nongenic factors are important does indicate that nongenic factors may be of importance also in situs inversus. There is no case on record of situs inversus being associated with virus infection of the mother. Virus infection has been reported to cause the syndrome of aberrant right subclavian artery, an anomaly of symmetry which accord-

ing to Brean and Neuhauser, has been seen in association with transposition.

The development of the asymmetry of the viscera and of the heart cannot be considered as an isolated process, the growth of the body taking place simultaneously. As far as I can see neglect to take this fact into consideration is the weakness of the theories of asymmetry reversal and transposition of the large vessels.

The influence of the growth differentials of the spine, lung buds and branchial arches indicates that the common mechanism of many cases of situs inversus is abnormal local growth rates. The vessels differentiate in situ, and in case of abnormal growth, in abnormal places. The joining of the vessels with the heart depends on their plasticity, which, according to the variability of the vessels, is great. In early stages a rearranging of the material during growth, in other words regulation or regeneration, may play a part tending to compensate for the abnormal growth rate. This may partly be the reason why so few of the persons with situs inversus show associated anomalies. These are more common in persons with heart defects, probably because the disturbances in the latter take place during a longer period. The abnormal growth concerns in most cases transitory structures, such as the gill arches, a fact explaining why the associated abnormalities tend to be transitory and why the anomalies of the heart so often have the character of atavistic phenomena.

The data given in foregoing pages indicate that abnormal symmetry or asymmetry of the vessels of the gill arches and of the aortic branches may produce either complete or partial inversion, particularly in the heart. Abnormal growth rates of the lung buds may cause abnormal position of the pulmonary vessels. The result will mostly be complete situs inversus, owing to the fact that the lung buds are related both to the foregut and to the heart. The other forms of partial inversion may be due partly to disturbances of growth rates of other parts of the body, partly to anomalies of the arteries of the branchial arches. The latter may extend their influences far caudally because of the caudal migration of the heart. Thus Pernkopf (1926) found symmetric pulmonary lobes in a case of isolated inversion in the abdominal cavity. A disturbance of the asymmetry of the organs of the septum transversum may cause inversion and anomalies of the heart from the caudal end as in the case showing inversion of the gallbladder and heart defect. The organs of the septum transversum and the derivatives of the midgut have a rather independent position in asymmetry. The diaphragm is often the border between a caudal and a cranial inversion. The stomach, duodenum, liver, gallbladder and large bowel may show isolated inversion and abnormal symmetry both in topography and in structure. The abnormal spleen in the person

with situs inversus and heart defect is probably due to abnormal asymmetry of the dorsal mesentery. Toldt (1889) attached importance to the symmetry of the dorsal mesentery in the development of the spleen.

E. Ludwig and Pernkopf (1926) took it for granted that bilateral organs, such as the lungs, have no influence on the situs viscerum. The evidence of the present study indicates the opposite, that the situs of the viscera depend on bilateral growth differentials of the viscera and the body and corresponding changes in the position of the vessels. Probably for this reason the genic mechanism of bilateral expression of the genes and that of asymmetry reversal of the viscera show striking similarities.

The conception that the embryonic vascular bed is an important factor of heart defects is not new. Taussig (1947), referring to Streeter, came to the same conclusion. According to her experience, the anomaly most commonly associated with heart defects is malformation of the spine, a finding which conforms with the observations concerning situs inversus.

The correlation of growth of the body and defect of the heart is evident in cases of mongolism and of arachnodactyly. According to Abbot, transposition has been recorded in cases of mongolism. That there is an influence on the asymmetry of the viscera in these cases is evident from the frequent symmetries of the lobes of the lungs.

I have not found any data showing that asymmetries of the body are commonly associated with heart defects. According to Brown, supernumerary nipple is one of the most common anomalies in cases of heart defects. This observation is of interest in view of the observation of Landauer (1939) that there is a correlation between an abnormality of symmetry, such as left-handedness, and supernumerary nipple.

Huxley, discussing relative growth, defines as instances of accretionary growth cases in which the material which is added is not alive. Such growth processes may result in spirals. In a study of the comparative anatomy of the stomach (Torgersen, 1942) I came to the conclusion that accretionary growth in this sense represents a particular case of a more general morphogenetic process concerning cases in which layers have different rates of growth. The layer showing the slowest rate has an influence on the form similar to that of the dead material.

The transposition of the large vessels and the possibility that this anomaly is related to visceral inversion have called on the interest of some of the most brilliant authors in different fields of biology and medicine (Harris and Farber).

To proceed from the cranial part, abnormal development of the mandibular arch and of the embedded vessels may cause complete asymmetry reversal and cardiac defect; a right subclavian artery from the left side

of the aorta may be associated with transposition. In the tetralogy of Fallot, in which transposition is one of the abnormalities and which is often combined with complete or partial inversion, particularly of the heart, a right-sided aortic arch, a mirror image of the normal, is found in 25 per cent of the cases.

Genic and nongenic factors may be supposed to affect the growth of the metameres or of the branchial arches, the most rapidly growing parts in different stages of development being most sensitive to growth-inhibiting influences. This causes abnormal asymmetry of the vessels in the corresponding regions. The latter condition puts a stress on the developing heart tube resulting in abnormal asymmetry of the bulbus and the common arterial trunk. This abnormal asymmetry affects the torsion of the truncus-bulbus septum and the bulboventricular loop in a different way. The truncus and the cranial part of the bulbus develop in a relatively fixed mesodermal sheet. Both Spitzer and Pernkopf and Wirtinger attach importance to the fixation of both ends of the cardiac tube. The bulboventricular loop is loosely fixed. The inner layer is growing faster than the outer layer, expanding the latter. In the relatively fixed truncus the torsion can take place only between the layers, manifesting itself in the truncal septum and the bulbar ridge. The process corresponds to that which, according to Jacobshagen (1931, 1934), takes place in the development of the spiral valve in the fixed mesodermal sheet in the spiral intestine of fish. The part of the bulboventricular loop that is not fixed undergoes a relatively free topographic torsion in a way which provides for a meeting of the septums. The explanation of the precision of these processes, according to this theory, is that they are different manifestations of the same causes. Abnormal asymmetric or symmetric influences may produce the different types of transposition with or without inversion.

This theory is similar to that of Pernkopf and Wirtinger. According to these authors who based their reasoning on embryologic evidence, a partial inversion is the cause of transposition. Spitzer and Pernkopf agreed that partial inversion may occur in different sections of the heart.

The bulboventricular loop does not undergo an entirely free torsion. In relation to the small intestine the heart tube is relatively fixed in the same way as the stomach. This is expressed in the muscular structure of both heart and stomach. Taussig (1926) and Pernkopf (1926) found that the superficial layer was not reversed, in contrast to the middle layer, which showed a mirror image asymmetry. The structure of the musculature and the formation of the vortex are influenced by the asymmetry reversal. Thus the torsion which depends on the strain put on the heart tube by the embryonic vessels produces at the same time the orientation of the septums and the organization of

the musculature which are fundamental in hemodynamics. In later stages the blood current may be assumed to be essential in the normal and in the abnormal development of the heart. These data are good evidence that the modifiers concerned with asymmetry are fundamental not only in the topography of the viscera but in the structure and in the normal and the pathologic function as well. This is confirmed by the observations of the anatomy of the stomach, which will be dealt with in subsequent paragraphs.

The torsion of the mesentery is an example of free torsion without any influence on the structure of the organ. There is no fundamental difference between the torsion of the mesentery and those of the bulboventricular loop, the stomach, the bulbus-truncus septum and the spiral valve. This is indicated in the fact that abnormal torsion of the mesentery and abnormal symmetries in the abdominal cavity are commonly associated with situs inversus and cardiac defects. These observations and the high correlation of the most different anomalies of the heart and situs inversus are good evidence that in cardiac defects the fundamental process is a disturbance of symmetry.

There are a few cases on record indicating a hereditary relationship between situs inversus and cardiac defect, among them the cases reported by Pernkopf (1937) and Roesler. The latter reported a case in which transposition occurred with partial inversion of the heart in one brother and clinical congenital heart disease in another. The parents were first cousins. The observations of familial occurrence of heart defects are convincing evidence of the influence of genic factors. The high correlation of heart defect and visceral inversion is evidence that the modifiers concerned with asymmetry are of importance in the mechanism of inheritance of heart defects. This does not exclude the influence of single genes.

The studies of Pernkopf (1926, 1937) and data in the literature show that the viscera in situs inversus may have abnormal shape and structure. This may have some interest, owing to the predisposition toward ileus in abnormalities of the mesentery.

The peculiar shape of the stomach of the twin showing isolated abdominal inversion is of particular interest. Pernkopf (1926) found the same shape of the stomach in 2 cases of isolated inversion of this organ. What surprised him was the observation that there was no pyloric sphincter in either of these two stomachs, one of them not even showing a pyloric sulcus. The pyloric part of the stomach looked like a part of the intestine. In a third of these extremely rare cases he found a strong pyloric musculature, looking like a case of pyloric spasm. This case showed the unique combination of an inverted stomach and a normal, not inverted, duodenum. For this reason the transition from the pyloric part of the stomach to the duodenum showed

a sharp angle. In the 2 first cases and possibly in the case in the present study the malformation was in a way the counterpoint to the hypertrophy observed in infantile pyloric stenosis and has, as far as I know, not been observed in the normal situs. In the study of the stomach I found that the growth differentials of the curvatures and the relative fixation of the stomach toward the transition to the midgut may be supposed to be of particular importance to the development of the pyloric musculature. The modifiers concerned with asymmetry are of importance in this fixation of the caudal part of the stomach, in the same way as they are essential in the fixation of the mesentery generally. The case in which the stomach was inverted and the duodenum not inverted confirms these suppositions, the pyloric part being abnormally fixed by the peculiar position of the duodenum.

It is evident that infantile pyloric stenosis may have a genic basis. The attempts to demonstrate a "single factor" mechanism have not been convincing. There is reason to assume that the modifiers which are fundamental in asymmetry are fundamental also in this disease. These children show in a way an exaggerated development of a gastric muscular structure which depends on the asymmetry of the organ. In contrast with children presenting congenital anomalies, they have been found in most cases to be taller than the average when examined as "grown ups" (Salmi). Like the patients with ulcer, they belong mostly to the slender type. Concerning the alimentary diseases observed in the families of children with pyloric stenosis, Cockayne and Penrose have supposed that some of these diseases may be manifestations of the gene in the heterozygous form. It might be added that the conformity noted as to sex ratio and body build in cases of ulcer of the stomach and duodenum and cases of infantile pyloric stenosis indicates a common genic basis also concerning the modifiers.

Complete situs inversus was probably about equally frequent in the sexes. In cases of persistent truncus arteriosus and cases of transposition taken together there are, according to White and Mönckeberg, 72 per cent boys; in cases of pyloric stenosis there are 80 per cent boys. There is reason to assume a sex difference with regard to the modifiers of the development of asymmetry and the related modifiers of the development of the particular organs. Conditions indicating a high degree of asymmetry or of abnormal symmetry are more common in males. According to Cummins and Midlo, a decrease of the bilateral asymmetry of the fingerprints is seen in left-handed males, and increase in left-handed females, the net effect being a leveling of the sex distribution of bilateral asymmetry, females being more symmetric generally. Left handedness is more common in boys. The correlations between this anomaly and supernumerary nipples and between the latter and heart defects are evidence of a relationship between sex, symmetry and

heart defects, a strong gynic component tending to produce abnormal symmetry in males. The similarity as to the influence of sex in different groups of asymmetries is good evidence of a similarity as to the genic mechanism. Sex influences the expression of the genes in much the same way as laterality does, both factors being of importance also in the expression of genic factors having relationship to the asymmetry of the viscera. Perhaps the most reasonable interpretation may be that the autosomes, which are supposed to have a masculine effect, also have a prominent effect on growth and bilateral growth differentials. These effects may be supposed to be modified by the X chromosomes, which may be assumed to have an opposite effect concerning all three features.

Dunn and Landauer (1934, 1936) suggested that the different expression of the tail mutations in fowl is due to modifiers that have been accumulated for the sake of developmental safety. This conception is reasonable also in regard to the modifiers concerned in visceral asymmetry. The essential point of Spitzer's theory of transposition is that the septation of the heart and the development of the lungs and the pulmonary artery were necessary adaptations to terrestrial life and that the torsion of the heart tube has played a part in this development. Keith (1913) attached importance to the regression of the bulbus in accounting for pulmonary stenosis. The regression of the bulbus depends on the asymmetric growth of the bulboventricular loop. Accordin to Saphir and Lev, the regression of the bulbus is important also in transposition. Keith stressed the phylogenetic correlation of the bulbus and the gills in an anatomic comparison parallel to the observations and interpretations of the present study.

Spitzer considered the heart in transposition as a reptilian heart in man. He expressed the opinion that transposition has phylogenetic causes, visceral inversion ontogenetic causes. Transposition is an expression of a disturbance of what he called a "metastable equilibrium." Has this metaphor from physics any biologic sense? The reappearance of a reptilian heart in man, even in details, may indicate that the mutations and selection concerning the heart in phylogeny have represented a continuous process giving discontinuous results. At a particular step in the accumulation of modifiers fundamental changes may have occurred in the reactive potency of the embryo. The genic complex in the development of the heart may be in a metastable equilibrium, mutations making the heart recoil on one of the discontinuous and decisive steps in its evolution. The observations indicate that the genic complex in the asymmetry of the viscera has been important in these processes. They do not confirm Spitzer's postulation that transposition and inversion are quite different phenomena. The objection to Spitzer's theory that a reptilian heart is not seen in human ontogeny may be regarded as irrelevant. In the evolution of the heart bilateral growth differentials

and alternative variability have played a great part. The alternative that is not realized leaves no trace is normal ontogeny. It reveals its presence as a possibility in the abnormal development of the heart.

SUMMARY

This study is based on 168 cases of situs inversus viscerum observed in Norway. They were partly detected in a mass roentgenography series comprising 1,000,000 persons, one third of the population. It is regarded as an advantage that the cases were collected from a small and geographically limited population. From the point of view of genetics all the cases must be regarded as selected. From the point of view of pathology they are partly selected, about one half of them having been reported by physicians. In all about 66 per cent of the persons with situs inversus in the country have been detected. The possibility that a very few involved siblings have not been detected is compensated for by the inclusion of two familial cases recorded by other authors. The plan of the study has been to make an approach by means of this abnormality of symmetry to the part played by the factors of visceral asymmetry in the development and the pathologic changes of the viscera, particularly the heart, the lungs and the stomach.

In Norway the frequency of situs inversus is probably somewhat above 0.01 per cent. There is evidence of geographic variations which parallel the variations of the frequency of twinning. The causes of these variations are discussed.

The frequency of situs inversus is not much higher in monozygotic twins than in the population. In some of the few families in which many twins occurred, the patient with situs inversus was a twin, a fact indicating a relationship between twinning and this anomaly in a few cases.

Situs inversus is probably rare in dizygotic twins. Among the cases in the present study is one in which the anomaly was found in both of a pair of dizygotic twins. The theoretic aspect of situs inversus occurring in dizygotic twins is discussed.

The age of the mothers is probably high in the cases complicated with malformations of the heart or with bronchiectasis.

The ratio of affected to normal persons does not accord with the supposition that a single recessive gene is generally the cause of situs inversus. In the present material there is no evidence of an increased prenatal selection or of unknown phenotypes which may explain why the actual ratio falls short of the expected ratio in recessive inheritance.

The low frequency of marriages of first cousins in the familial cases and the lack of involved siblings in cases in which marriages of first cousins were noted are difficult to match with the supposition of a single recessive gene.

Situs inversus is more equally distributed between the sexes than heart defects and transposition of the great vessels. The genic basis of the correlation between asymmetry, general growth and sexual differentiation in different groups of asymmetries in man is discussed.

Anomalies of the spine are increased in persons with situs inversus, as well as defective development of the lower jaw. This may indicate inductive relationships or an influence of abnormal growth rates. These findings are good evidence that the growth rates of the gills and the metameres and accordingly the position of the embedded vessels are of importance in many cases of asymmetry reversal and heart defects.

The relationship between situs inversus and heart malformation may be regarded as a clue to an understanding of the developmental processes and causes of congenital heart disease. On this basis a theory is proposed concerning the transposition of the large vessels, including the main points in the previous theories of Pernkopf, Wirtinger, Spitzer and Keith.

Bronchiectasis occurred in about 25 per cent of the cases and in 4 of the 5 familial cases. This frequency of bronchiectasis and nasal polyps is discussed, together with the difficulties of their diagnosis. Bronchiectasis and nasal polyps may occur as different manifestations of a common genic basis. The frequent coincidence of nasal polyps and bronchiectasis and the fact that the frontal sinuses are small in cases of situs inversus complicated with bronchiectasis and not in the other cases may be regarded as good evidence of a common genic basis of bronchiectasis and nasal polyps. The syndrome behaves as a dominant with varying expression and chance of inversion in heterozygotes and homozygotes. This combination may be a clue to the understanding of the relationship between genic factors in the morphology and the asymmetry of particular organs and the asymmetry of the viscera. Evidence from morphology and genetics indicates that the asymmetry of the viscera is due to a complex of genes which may be broken up into components of particular importance in particular organs. The complete asymmetry depends on the integrative action of this complex of genes. The role of these modifiers in normal asymmetry in the morphologic and pathologic aspects of the viscera is discussed with particular regard to the lungs, the heart and the stomach. A theory is proposed concerning the influence of these modifiers in infantile pyloric stenosis. The genic aspects of the phylogenetic theory of Spitzer concerning transposition are discussed.

ADDENDUM

Since the manuscript was submitted for publication the material has increased substantially. It now includes 185 families and 195 cases of situs inversus. About 1,500,000 persons have been examined

by mass roentgenography. In the following paragraphs are recorded some data of interest in the present study.

Twin birth occurred in 1.7 per cent of 1,082 births in the sibships. Of 57 families, one of the grandparents was a twin in 3, and one of the parents in 2—corresponding to a frequency of 1.3 and 1.7 per cent respectively. In a sibship of 9 situs inversus occurred in 2 siblings. The father was a twin. Situs inversus has not been detected among the siblings in the cases recorded. Two more familial cases have been detected. Besides the family in which the father was a twin, a sibship was detected in which 3 siblings showed situs inversus and 1 the normal situs. The parents were second cousins. The lung-nose syndrome occurred in both these families, all the 5 persons with situs inversus showing symptoms of bronchiectasis, 2 of them nasal polyps as well. The mean age of the mothers in 191 cases is 31.4. The difference from the age of mothers at birth generally is 0.9 ± 0.43. The numbers of affected and normal siblings, the index cases excluded, are 10 and 897 respectively. The frequency of first cousin marriages is 3.2 per cent, of second cousin marriages 7.7 per cent. In families showing the lung-nose syndrome the frequency of first cousin marriages is 6 per cent and of second cousin marriages 16 per cent. In the other families the corresponding frequencies are 2.2 per cent and 3.8 per cent. The difference between the groups concerning consanguineous marriages is 16 ± 7.2. No situs inversus has been detected in 39 siblings in the 6 families in which the parents were first cousins. In 45 families showing the lung-nose syndrome the frequency of situs inversus in the siblings was 5 per cent, against 0.16 per cent in the siblings in the other families.

The supplementary data confirm the interpretations in the preceding pages. The data are evidence of an interaction between genes affecting the particular organ or regions connected with it and a genic complex which gives response to selective breeding and influences the growth differentials determining the asymmetry of the viscera.

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EFFECT OF TRIPELENNAMINE HYDROCHLORIDE ON BURN SHOCK

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SINCE the work of Dale and Laidlaw,[1] it has been recognized that histamine or a histamine-like substance may be responsible in part for the development of shock. The evidence in support of this theory has, however, been conflicting. Moon [2] and Harkins [3] have recently presented reviews on this subject. The discovery of drugs, such as pyranisamine (neoantergan,® [N,N-dimethyl-N'-(p-methoxybenzyl)-N'-(alpha-pyridyl) ethylenediamine]), diphenhydramine (benadryl® [beta dimethylaminoethyl benzhydryl ether]), tripelennamine (pyribenzamine® [N,N-dimethyl-N'-Benzyl-N'-(alpha-pyridyl) ethylenediamine]) and others, which have the properties of histamine antagonists, suggested a new means of investigating the role played by histamine in shock. Friedlander, Feinberg and Feinberg,[4] and Sherrod, Loew and Schloemer [5] have shown that tripelennamine will prevent shock from developing after intravenous injection of histamine in guinea pigs and dogs. However, Jourdan and Chatonnet [6] and Ingraham and Wiggers [7] have reported that pyranisamine and diphenhydramine do not alter the course of either traumatic or hemorrhagic shock. The purpose of the experiments to be described was to extend the work of these authors and to study the effects of tripelennamine in burn shock.

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METHODS AND MATERIALS

Fourteen female albino rabbits were used in this experiment. These animals had previously been used for the Friedman test but were in good health, and the incisions were well healed. Seven animals were used as controls and 7 received tripelennamine.[8] The treated group were given 4 mg. of tripelennamine hydrochloride per kilogram of body weight. The drug was dissolved in isotonic solution of sodium chloride and injected into the marginal vein of an ear. These animals were given 1 mg. of the drug per kilogram every two hours until the conclusion of the experiment. Ether anesthesia was used to maintain unconsciousness during the period of burning and for a few minutes thereafter. Animals 1 and 8 were clipped, but the fur of the remaining animals was soaked thoroughly with water to insure even heat conduction before burning. A control blood sample

Comparison of the Hematocrit Readings Made for Rabbits Treated with 4 Mg. of Tripelennamine Hydrochloride per Kilogram of Body Weight and Untreated Rabbits (Controls), All of Which Were Immersed in Water at 80 C. for Ten Seconds

Animal	Hours After Burn													Cause of Death	Survival, Hr.
	0	2	4	6	8	10	12	14	16	18	20	22	24		
Treated															
1	41	48*	45*	..	28	..	..	..	..	..	..	..	..	Shock	8
2	39	39	36	..	..	..	..	..	..	..	..	..	..	Tripelennamine?	5
3	49	50	..	..	..	..	..	..	..	..	..	..	..	Tripelennamine?	2
4	42	47*	47*	..	..	..	..	..	..	..	..	..	..	Shock	4
5	44	41	54*	..	50*	..	42	..	46	..	41	..	..	Convulsions	25
6	35	45*	..	42*	..	..	..	..	..	..	..	..	..	Hemopericardium	7
7	38	57*	49*	51*	..	50*	..	..	..	..	..	..	..	Shock	12
Untreated															
8	43	41	51*	..	47*	..	48*	..	47*	..	46*	..	40	Put to death	27
9	42	53*	51*	..	55*	58*	..	..	..	..	..	..	..	Shock	10
10	42	34	51*	..	50*	..	47*	..	47*	..	46*	..	44	Hemopericardium	27
11	48	56*	56*	56*	..	48*	..	45*	43	..	..	..	36	Hemopericardium	23
12	32	32	29	..	30*	..	37*	..	32	..	30	..	..	?	40
13	44	16?	54*	..	56*	..	52*	..	48*	..	49*	..	48	Put to death	52
14	40	42*	44*	..	45*	..	40	28	..	..	38	..	36	Pneumonia	36

* The animal was in shock.

was drawn from each rabbit by cardiac puncture, and the animals were then immersed to the xiphoid in water at 80 C. for ten seconds. The rabbits were placed in cages and fed a commercial rabbit chow and as much water as they would drink.

Hematocrit determinations were made for all animals at two to four hour intervals, and for 8 animals erythrocyte counts were also made. Blood samples were drawn by cardiac puncture. Survival time was noted, and complete autopsy was done immediately after death.

RESULTS

Twelve of the 14 animals in this series underwent shock, which was recognized by an increase in the hematocrit value and in the red blood

8. Pyribenzamine hydrochloride,® supplied by Ciba Pharmaceutical Products, Inc., was used.

cell count, by pallor of the ear, coldness and lethargy. The pulse and respiratory rates were too variable to be used for analysis. Two of the animals in each series were burned for twenty seconds, but the results were not altered by this increase in length of burning. Animals dying of hemopericardium and those dying before the onset of shock were eliminated from the studies of longevity.

Of the 7 animals which received tripelennamine, 5 showed evidences of shock. Two animals died within five hours of burning without revealing hemoconcentration and were therefore eliminated from the series. Four animals died in shock. One animal died of hemopericardium after recovering from shock. The average length of life in this group was ten hours, and all except 1 animal died within four to twelve hours.

In the control series all 7 animals presented evidences of shock, but 6 of them recovered from the shock before death. One animal died of hemopericardium while in shock. Two were killed after recovery to avoid infection of the burned areas; 1 died of hemopericardium, and 1 of pneumonia after recovering from shock. The average survival time for this group was thirty-three hours, with all but 1 living twenty-seven to fifty-two hours.

Of the 5 shocked animals in the treated series, 4 showed first signs of hemoconcentration at two hours and 1 at four hours. In the control series, hemoconcentration tended to occur later, with 3 showing first signs of increased cell volume at two hours, 3 at four hours and 1 at eight hours.

The highest hematocrit reading was 57 in the treated group and 56 in the control group, the average hematocrit peak being 50 in both groups. The peak hematocrit values tended to occur later in the control group. The average maximum increase of the control group was 23 per cent and that of the treated group 26 per cent. The range of percentage increase was 12.5 to 31 in the control group, and 12 to 50 in the treated group.

The erythrocyte counts checked with the hematocrit readings. The changes observed at autopsy in one series of animals were similar to those observed in the other. Shock was accompanied by pulmonary edema, hyperemia of the viscera and engorgement of the veins of the abdomen.

COMMENT

In 1939 Wense[9] induced burn shock in normal animals and in animals desensitized to histamine or animals previously given torantil[9] (desiccated kidney and extract of the mucous membrane of the small intestine of the hog, containing histaminase) and found no difference in

9. Wense, T.: Ztschr. f. Immunitätsforsch. u. exper. Therap. **97**:100, 1939.

the survival time but thought the local reaction less in the treated animals than in the control animals. If histamine were the "burn toxin" producing the vascular collapse and shock after severe burns, it would seem that tripelennamine, if an effective antagonist of histamine, should alter the course of events after a standard burn. However, in these studies no decrease of severity, delay of onset or inhibition of progression of shock was noted. The only significant change was decreased viability of the animals given tripelennamine hydrochloride. The drug, therefore, seems to exert no effect on the mechanism of burn shock and acts only as an additional toxic factor in an already traumatized animal.

SUMMARY

Burn shock was induced in 12 of 14 rabbits. Seven of the animals received large doses of tripelennamine hydrochloride intravenously.

No decrease of severity, delay of onset or inhibition of progression of shock was noted in the treated series.

The treated series showed decreased viability.

PRIMARY SYSTEMIC AMYLOID DISEASE

Report of a Case Emphasizing Cardiac Involvement

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AMYLOID is a peculiar member of the group of connective tissue hyalins, set apart from the others by certain identifying characteristics. These are its staining reactions,[1a] its appearance in persons who have passed through a long wasting illness [1b] and the fact that it involves principally the parenchymatous organs (liver, spleen and kidneys) and the adrenal glands.[1c] Lubarsch [2] is recognized as the first to point out cases in which a hyaline eosinophilic substance, possessed of certain of the morphologic properties of amyloid and conspicuously lacking others, makes its appearance in tissue. These cases are designated as instances of "primary systemic amyloidosis"; they are recognized by criteria initially formulated by Lubarsch and employed by subsequent writers.[3] In what is deemed the order of their importance, these are: (*a*) absence of such a predisposing factor as one of the chronic suppurations; (*b*) failure of the amyloid deposit to show the usual staining reactions; (*c*) no involvement of the organs commonly affected by secondary amyloidosis; (*d*) extensive amyloidosis of such sites as the heart, blood vessels, skin and skeletal muscle not commonly involved in secondary amyloidosis.[3a] In these cases the infiltrate resembles that seen in amyloidosis secondary to pulmonary tuberculosis, chronic osteomyelitis, infected tumors, leprosy, chronic nephritis and other diseases [1a] in its gross appearance and in its reaction to the routine histologic stains. Its response to the special stains for amyloid, i. e., dilute Lugol's solution and sulfuric acid, congo red and crystal violet,

1. (*a*) Karsner, H. T.: Human Pathology, Philadelphia, J. B. Lippincott Company, 1942. (*b*) MacCallum, W. G.: A Textbook of Pathology, Philadelphia, W. B. Saunders Company, 1936. (*c*) Smith, L. W., and Gault, E. S.: Essentials of Pathology, New York, D. Appleton-Century Company, Inc., 1942.

2. Lubarsch, O.: Virchows Arch. f. path. Anat. **271**:867, 1929.

3. (*a*) Perla, D., and Gross, H.: Am. J. Path. **11**:93, 1935. (*b*) Reiman, H. A.; Koucky, R. F., and Eklund, C. M.: ibid. **11**:977, 1935. (*c*) Koletsky, S., and Stecher, R. M.: Arch. Path. **27**:267, 1939. (*d*) Lindsay, S., and Knorp, W. F.: ibid. **39**:315, 1945.

is irregular and capricious.[4] In some cases the material shows an affinity for one or more of these dyes; in others it fails to do so. Even in tissues from the same patient, areas otherwise morphologically identical may fail to show the same reaction to these tests.[5] This phenomenon has suggested the existence of a preamyloid stage of development[6] or the presence of multiple types of protein which are being identified collectively as amyloid.[5]

To date, some 50 cases of primary systemic amyloidosis have been recorded in the literature. On 48 of these autopsy reports are available. Koletsky and Stecher[3c] and Lindsay and Knorp[3d] identified, discussed and tabulated the findings in 40 cases reported up to 1945. In 1946 Lindsay[7] discussed 4 additional cases, including 2 which had previously been overlooked, and added 1 of his own. During and subsequent to 1946 reports of 5 additional cases, not included in the foregoing reviews, have appeared in the literature.[8] The frequency with which such reports have been made during the past three years indicates that this disease may not be quite so rare as was previously thought. An additional case is now reported.

REPORT OF CASE

A 39-year old married white woman entered Oliver General Hospital on Feb. 5, 1948 and died eighteen days later. On admission to the hospital she complained of weakness of the right arm and the right leg. She stated that eight days before admission, after retiring for the night, she suddenly found herself unable to speak; her right extremities were limp and could not be moved. She regained her speech the following day. Use of the extremities gradually returned so that at the time of admission she could walk without difficulty, although the arm and leg were still weak.

The family history was noncontributory. The father, the mother and ten siblings were all living and in good health. One sister died of uterine cancer at the age of 47.

Except for typhoid fever at the age of 6, the patient recalled no significant illnesses until 1936, when the last of six children was born. This child was stillborn, and the labor lasted about forty-two hours, terminated by forceps. On the ninth postpartum day thoracic pains of pleuritic type developed, with fever and cough. This illness followed a protracted course, with exacerbation of the thoracic pain after the second week and swelling of both legs after the third week. She was hospitalized for one month and remained in bed at rest for a second month.

4. (*a*) Kerwin, A. V.: J. Lab. & Clin. Med. **22**:255, 1936. (*b*) Eisen, H. N.: Am. J. Med. **1**:144, 1946. (*c*) Golden, A.: Arch. Int. Med. **75**:413, 1945.

5. Iverson, L., and Morrison, A. B.: Arch. Path. **45**:1, 1948.

6. Eklund, C.: Bull. Staff Univ. Minnesota Hosp. **5**:180, 1934; cited by Iverson and Morrison.[5]

7. Lindsay, S.: Am. Heart J. **32**:419, 1946.

8. (*a*) Orloff, V., and Felder, L.: Am. J. M. Sc. **212**:275, 1946. (*b*) Lindsay, S.: Am. J. Med. **4**:765, 1948. (*c*) Golden.[4c] (*d*) Iverson and Morrison.[5]

Following this illness, she continued to have some intermittent swelling of the feet on prolonged standing, weakness, nervousness and fainting spells; however, she was able to hold a strenuous job in an aircraft factory for several years during the war.

In 1942 she began to have epigastric aching related to meals, relieved by soda, soft drinks and food. This continued until the time of admission, with some exacerbation during the year prior to admission. For two years there had been gradually increasing dyspnea on exertion, paroxysmal nocturnal dyspnea, orthopnea and edema of the legs, which at the time of admission extended above the knees. One year prior to admission she was told by a physician that her heart and liver were enlarged. For the last four months there was an intermittent pain of cutting type in the right upper quadrant of the abdomen. For three months she had noted that her abdomen was enlarging although she was losing weight. There had been a total loss of about 50 to 60 pounds (22.5 to 27 Kg.) in the one and one half year period preceding admission.

Examination revealed a well developed 39 year old white woman with pale, ashen complexion. She was apathetic, but appeared in no acute distress. The skin of her feet, ankles and wrists was thickened, scaly, slightly reddened and tender to pressure. There was pitting edema to the middle of both legs. There was marked gingivitis. The tongue was beefy red; motility was normal. There were coarse crepitant rales in both pulmonary bases posteriorly. Her heart was enlarged to the left; the tones were distinct, but no murmurs were heard. The blood pressure in the right arm was 105 systolic and 80 diastolic; that in the left arm, 108 systolic and 76 diastolic. There were infrequent premature beats. The circulation time, arm to tongue, with sodium dehydrocholate, was 27 seconds. The venous pressure was 23 cm. of isotonic solution of sodium chloride. The abdomen was protuberant in contrast to the wasting of the remainder of the body. A distinct, sharp edge of the liver, hard, apparently nodular and nontender, was palpable below the level of the umbilicus. The abdomen was distended and tympanitic. The spleen was not palpable. The neurologic examination revealed a hyperactive right knee jerk; the remainder of the peripheral reflexes were hypoactive (but present). Babinski, Oppenheim and Romberg signs were not present. There was no evidence that cranial nerves were involved. The muscular strength of the arms and legs was equal bilaterally.

Urinalyses showed leukocytes intermittently with occasional erythrocytes; the albumin content varied from none to 2 plus. The white blood corpuscle count on admission was 18,400, with neutrophilic granulocytes 71, lymphocytes 27 and eosinophilic granulocytes 2 per cent; it varied from 10,000 to 17,000 during the hospital course, with a moderate shift to the left.

Other laboratory results were:

Hematocrit reading, 42 per cent
Sedimentation rate, 16 mm. per hour (Wintrobe, corrected)
Prothrombin concentration, 100 per cent
Cephalin-cholesterol flocculation (Hanger's test), negative
Thymol turbidity test, negative
Quantitative van den Bergh test—1 minute 0.2 mg., total 0.5 mg., per hundred cubic centimeters of serum
Blood proteins

Albumin 3.23 Gm.	per hundred cubic centimeters
Globulin 2.15 Gm.	
Total 5.38 Gm.	

Blood urea nitrogen, 11 mg. per hundred cubic centimeters
Spinal fluid
Sugar 50 mg.
Protein 15 mg. } per hundred cubic centimeters
Chlorides 700 mg.
Wassermann and colloidal gold tests, negative

Repeated electrocardiograms showed low voltage QRS complexes and flattening to inversion of T waves in leads I, II and III. R waves were absent in CR 1 to 3, and T waves were inverted in CR 5 and 6 (fig. 1).

In roentgen studies, the heart was diffusely enlarged in the anterior-posterior view. In the oblique views there was some impingement on the retrocardiac space.

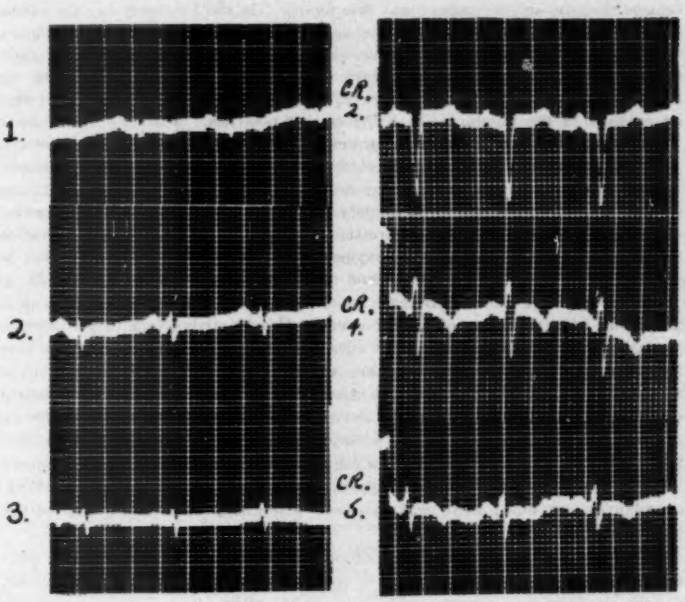

Fig. 1.—Electrocardiogram taken Feb. 10, 1948. Note the low voltage of QRS and flattening to inversion of T waves in limb leads. R is absent in CR2, and T waves are inverted in CR4 and 5.

Under fluoroscopic examination the amplitude of pulsations appeared somewhat diminished. The great vessels did not appear dilated. It was the impression of the radiologist that no great amount of fluid was present in the pericardial sac, but the presence of some fluid could not be excluded. The general picture appeared consistent with avitaminosis or, possibly, myxedema. The lower lung field showed some haziness, believed to be due to congestion. A gastrointestinal series showed only a dilated duodenal cap with a questionable pseudodiverticulum on the side of the greater curvature.

Course in Hospital.—The patient was placed in bed at rest and was supplied with a low salt, low fat, high carbohydrate, high protein diet with supplementary

vitamins consisting of 10 mg. of thiamine hydrochloride, 2 mg. of riboflavin, 100 mg. of nicotinamide, 100 mg. of ascorbic acid and one vitamin A-D capsule three times daily. She improved considerably during the first week in that her shortness of breath disappeared, as did the beefy redness of her tongue. The skin of the ankles and wrists became more normal in appearance, and the tenderness to pressure disappeared. On February 10 she was allowed bathroom privileges. During the following week she remained fairly comfortable, complaining mainly of nagging abdominal discomfort and occasional dizzy spells. There was no decrease in the hepatomegaly.

During the night of February 19 there suddenly developed massive edema of the lower extremities, the blood pressure fell to 90 systolic and 60 diastolic, the pulse became thready, and the patient was disoriented. On the following day the edema had decreased. She was semistuporous and showed a lag of the facial musculature with ptosis of the right eyelid. There was complete motor paralysis of the right arm and leg with hyperactive reflexes, ankle clonus and Babinski's great toe reflex on that side. A spinal tap revealed a pressure of 270 mm., rising to 350 mm. on pressure of the jugular veins. The cerebrospinal fluid was clear, with sugar 50 mg., chlorides 700 mg. and total protein 15 mg. per hundred cubic centimeters. A tentative diagnosis of thrombosis of the left middle cerebral artery was made.

The cardiac enlargement appeared somewhat increased, but a pericardial tap showed no evidence of fluid. The patient was digitalized without notable improvement. The following day mental depression increased and Cheyne-Stokes respiration supervened. She was placed in an oxygen tent but grew progressively weaker in spite of intensive supportive therapy and died during the night of February 23.

A necropsy was made thirty-three hours after death, after contact with next of kin. The body was that of a well developed, moderately obese white woman, appearing approximately 45 years of age. The abdominal wall was flabby, with numerous silvery gray cutaneous striae, especially in the lower quadrants. The right calf appeared slightly larger in diameter than the left; there was minimal edema of the extremities. Within the abdominal cavity, approximately 350 to 400 cc. of clear amber-colored fluid was present; in the left upper quadrant a thin brownish material had escaped from a ragged defect in the fundus of the stomach. The left pleural cavity contained approximately 300 cc. of thin, foul-smelling, chocolate brown fluid; there was extensive autolysis of the lower third of the esophagus.

The heart weighed 600 Gm. Its contour was globoid; the chambers on the right side of the heart were prominent. The epicardial surface was smooth throughout, and the coronary vessels were readily followed beneath the epicardium by inspection and palpation. Within the chambers of the heart, the most prominent alteration was seen in the left atrium (fig. 3*B*). Here the endocardium was diffusely thickened, numerous small semitranslucent grayish tan plaques approximately 0.2 cm. in diameter were slightly upraised above the surface of the endocardium. These were distributed diffusely over the entire endocardial surface; they were more prominent in the area immediately superior to the posterior cusp of the mitral valve. The wall of the left atrium appeared definitely thicker than that of the right, but it did not appear especially stiffened. The endocardium of the remaining cardiac chambers was essentially normal in gross appearance. The cardiac valves were thin, and no nodularity was noted in the leaflets. The pulmonic valve ring was mildly to moderately dilated; it measured 9.2 cm. in circumference. The measurements of the remaining valve rings in centimeters were as follows: mitral valve, 10.2; aortic valve, 7.8; tricuspid valve, 12.3. The right ventricular wall

measured 0.5 cm., the left ventricular wall 2.0 cm., in thickness. Sections through the myocardium revealed mild to moderate increase in consistency; the cut surface was of a uniform reddish brown color.

The lungs were increased in weight; the right weighed 525 Gm. and the left 450 Gm. The posterior aspect of the left lung had been extensively eroded and softened by gastric contents which had escaped through the esophageal perforation. With this exception, the pleural surfaces were smooth and glistening. Crepitus appeared mildly to moderately reduced throughout both lungs. On cut section a relatively dry surface was noted, showing the usual anthracotic markings. The pulmonary vessels were not unduly prominent; compression of the lungs produced no fluid from the parenchyma or exudate from the bronchi.

The spleen weighed 535 Gm.; the general contour and the appearance of the capsular surface were within normal limits; it was obviously enlarged, and its consistence was definitely increased. Cut section revealed a smooth, glossy, dull red surface on which the normal structural markings were obscured; scraping produced only minimal particles of pulp.

The liver weighed 4,200 Gm. The central portion of the liver was obviously softened and yellowish; about the periphery a more normal appearance persisted. Here the structural markings suggested chronic passive hyperemia; the central portion of the hepatic lobule was accentuated by deep brown markings, while the more peripheral portion was outlined in lighter yellow-brown, giving the appearance of the so-called "nutmeg" liver.

The autolysis of the stomach and the esophagus previously referred to involved the posterior aspect of the lower third of the esophagus and the upper fourth of the stomach along the greater curvature; the margins of these defects were softened, ragged and greenish black. In these areas of autolysis the blood vessels were not spared. There was no evidence of chronic peptic ulcer.

Each kidney revealed two or three areas of ischemic infarction; these presented the typical gross appearance, with softened yellow centers surrounded by a brighter red zone of reactive hyperemia. Vascular occlusion could not be grossly demonstrated. The right kidney presented complete reduplication of pelvis and ureter; the superior and inferior renal pelves were separated by an isthmus of renal parenchyma. The separate ureters opened into the bladder by distinct ureteral orifices, 0.7 cm. apart.

The brain weighed 1,360 Gm. Its general consistence was much diminished; there was a large area of well marked encephalomalacia involving the inferior surface of the left frontal and temporal lobes together with the basal ganglions on that side. The left middle cerebral artery was occluded by a thrombus extending from the internal carotid artery throughout its entire course. Similar thrombotic occlusion likewise involved the left anterior cerebral artery, extending as far rostrally as the anterior communicating artery. The left middle cerebral artery had a small fusiform dilatation adjacent to its origin from the carotid artery. Serial sectioning after two weeks' fixation in 4 per cent formaldehyde solution revealed severe softening of the entire left frontal lobe together with the basal ganglions on the left. The cerebellum, pons, brain stem and right cerebral hemisphere presented essentially normal consistency.

A single small area of hemorrhage, approximately 0.3 cm. in greatest diameter, was noted in the midportion of the pons.

The adrenal glands combined weighed 21 Gm.; their consistency was moderately increased, especially in view of the thirty hour lapse prior to autopsy. Transverse section revealed that the usual bright yellow outer cortical stripe had been replaced by dull grayish brown material.

Microscopic Observations.—Heart: Beneath the endocardium and the epicardium of the left atrium (fig. 2 *A*) there appeared bands and masses of amorphous or finely fibrillar material, which in sections stained with Harris' hematoxylin and eosin presented a hyaline appearance; sections stained with crystal violet revealed the reddish purple metachromatic staining characteristics of amyloid. This metachromatic staining reaction was by no means uniform throughout the subendocardial deposit; it appeared in patchy areas separated by other zones in which typical metachromasia was not noted. Within the myocardium, extensive similar amyloid deposits were detected as hyaline eosinophilic material in the hematoxylin-

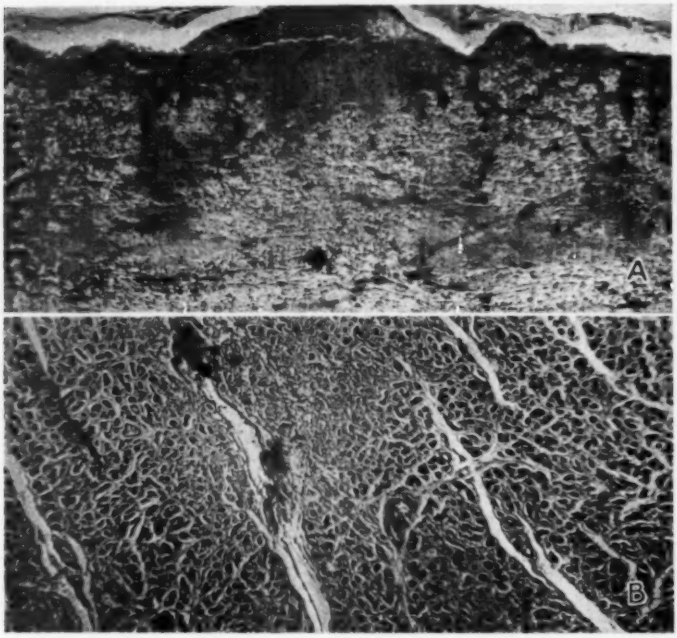

Fig. 2.—*A*, endocardium of the left atrium (× 100). Note the thickening of the endocardium and the irregular deposits of amyloid, which appears as denser material.

B, myocardium of the left ventricle (× 100). Note the thin amyloid rings about the muscle fibers; crystal violet stains show irregular metachromasia. Note the thickening of small arteries caused by presence of amyloid.

The photomicrographs in this and the following figure are from tissue fixed in 10 per cent formaldehyde solution, embedded in paraffin and stained with hematoxylin and eosin.

eosin section, a finding confirmed by crystal violet stains. Amyloid deposits varied from narrow rings about the individual muscle fibers (fig. 2 *B*) to larger strands and masses (figure 3 *A*) deposited between the muscle bundles. In its most extreme form the amyloid deposit totally replaced the muscular tissue. In the areas of

minimal deposition the histologic appearance suggested that the amyloid substance made its first appearance as a membrane-like deposit at the surface of the cardiac muscle fiber. In some areas the continuity of the muscle fibers appeared to have been interrupted by masses of amyloid, but it was impossible to determine that this appearance did not indicate that the muscle fibers had deviated from the plane of section rather than that the cytoplasm had been replaced by infiltrating amyloid. Prominent throughout all sections were amyloid deposits present within the media of the small branches of the coronary arteries. They appeared as small nodular masses between the muscle fibers or replaced them; in some instances, amyloid replacement had proceeded to such an extent as to occlude the lumen of the vessel. The amyloid deposited within the media of the arterial walls almost universally

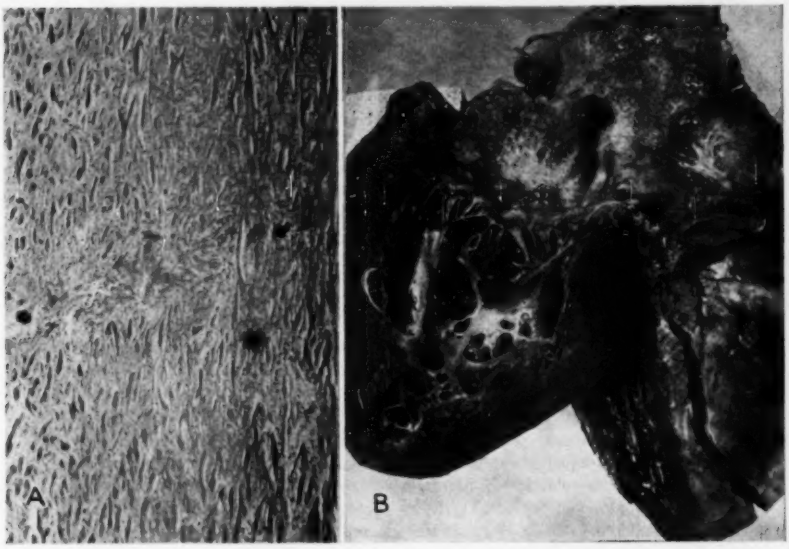

Fig. 3.—*A*, myocardium of the left ventricle (× 100). Massive intercellular deposits of amyloid are seen.

B, heart sectioned to show the left atrium and ventricle. The photograph is from the gross specimen, which has been preserved in Kaiserling's solution. Note the translucent subendocardial deposit in the atrial wall.

gave the metachromatic staining reaction with crystal violet. The infiltrating substance noted about and between the muscle fibers yielded a reaction similar to that observed beneath the atrial endocardium. Patchy areas revealed typical reddish purple metachromatic staining with crystal violet, while in adjacent zones this reaction failed to appear. Such irregular staining was seen alike in the areas of massive amyloid infiltration of the myocardium and in those zones of minimal deposit in which the substance appeared as a delicate layer on the surface of the muscle fibers. The deposition of amyloid was noted both within the small arteries lying beneath the epicardium and in those deep within the myocardium; certain

small branches of the cardiac veins displayed minimal deposition of amyloid within the adventitia. Examination of the subepicardial fat did not reveal pericellular deposits of amyloid.

Lungs: The smaller branches of the pulmonary artery and the pulmonary arterioles showed the media replaced to varying degrees by amorphous masses of hyaline, deeply eosinophilic material, which in sections stained with crystal violet failed to give the typical metachromatic staining reaction. Sections stained with phosphotungstic acid–hematoxylin differentiate this substance from collagen. In addition to this vascular infiltration small strandlike deposits of amyloid were noted within the interalveolar septums; in this location they paralleled the course of the precapillary arterioles and that of the capillaries. This material gave the staining reactions previously described. In several areas beneath the pleura, small triangular infarct-like areas of necrosis of the interalveolar septums were associated with intra-alveolar hemorrhage. Adjacent to such areas there was nearly always a small branch of the pulmonary artery in which the lumen had been completely obliterated by massive medial deposition of amyloid.

In addition to the amyloidosis described to this point there were small focal areas of bronchopneumonic infiltration of groups of alveoli adjacent to the bronchioles. Scattered "heart failure" cells were noted within some of the alveoli.

Spleen: The splenic parenchyma was almost totally replaced by masses of amyloid, which appeared to be deposited beneath the endothelium lining the splenic sinusoids. The sinusoids were markedly compressed, and the splenic cords were almost totally replaced. The malpighian corpuscles were so reduced in size and number that they were identified only with difficulty. Sections stained with crystal violet gave only a faintly metachromatic reaction; sections stained with phosphotungstic acid-hematoxylin revealed the infiltrating substance, stained reddish orange, which differentiated it sharply from normal connective tissue and muscle elements. The central arterioles of the splenic corpuscles revealed nodular deposits of amyloid replacing their media. The infiltrate here, in contrast to that noted within the splenic pulp, stained metachromatically with crystal violet.

Liver: Large amounts of amyloid material were deposited between the sinusoidal endothelium and the underlying liver cells; there was total obliteration of the normal hepatic structure in the central and midzones of the lobules; about the periphery a semblance of normal structure persisted. There was extensive medial amyloidosis of the small arteries throughout the section. Here, as in the spleen, the amyloid deposited within the walls of the blood vessels stained metachromatically with crystal violet, while that deposited beneath the sinusoidal endothelium failed to give this reaction.

Genitourinary System: The renal amyloid deposit was restricted to small nodular masses deposited beneath the endothelium of the glomerular capillaries; in no instance did this progress to complete obliteration of a glomerulus. The amyloid here failed to stain metachromatically. In contradistinction to the glomeruli, the renal vascular system showed fairly impressive medial amyloidosis of the arteries. Metachromatic staining was noted in some cases, while it was absent in others; vessels showing typical eosinophilic hyaline material in the section stained with hematoxylin and eosin but failing to show metachromasia in the section stained with crystal violet were usually the smaller branches of the renal artery. Sections taken through the margin of one of the grossly described infarcts revealed a typical histologic appearance with palely eosinophilic "ghosts" of renal structures. A small nodular deposit of amyloid was noted within the renal capsule in one section.

The uterine sections showed extensive amyloid replacement of the myometrium; the amyloid stained metachromatically. There was also medial amyloidosis in a few of the arteries of the myometrium.

Ovaries: Ovarian sections revealed amyloidosis confined to the media of the smaller arteries; metachromatic staining of the amyloid was noted.

Adrenal Gland: Sections of adrenal gland showed massive amyloidosis of the cortex; the change appeared most marked in the zona fasciculata, though less prominent changes were noted in the zona glomerulosa and the zona reticularis. In the cortex, the histologic structure suggested that the amyloid had been deposited beneath and between the epithelial cells of the adrenal cell cords. The epithelial cells were displaced into the sinusoids, with obliteration of the last-named structures in many instances. In medium and smaller-sized arteries beneath the adrenal capsule there was extensive medial deposition of amyloid. In the periadrenal fat the crystal violet stain gave a typical amyloid reaction in only small, scattered areas.

Central Nervous System: Sections of the brain showed the typical degenerative changes of extensive anemic softening in the areas grossly involved in the encephalomalacic process. Sections through the left midcerebral artery revealed occlusion due to a laminated thrombus of the antemortem type; section through the grossly described aneurysmal dilatation revealed arteriosclerotic changes within the wall of the artery. Phosphotungstic acid-hematoxylin revealed splitting of the internal elastic lamina with deposition of a fibrillar material having the staining affinities of collagen between the separated layers. There was also subintimal deposition of fibrillar amorphous substance which appeared to be atheromatous in nature. Certain sections revealed lamination of the vessel wall, the split occurring external to the internal elastic lamella. There is no evidence of amyloid deposition within the vessel wall.

Miscellaneous Sites: In addition to the deposits described, small deposits of amyloid were found in the media of the small and medium-sized arteries and arterioles of the duodenum, the ileum, the uterine tube, the pancreas and the bone marrow. The esophagus showed acute inflammation with edema of the wall and chronic inflammatory cell infiltration.

Pathologic Diagnoses.—Primary systemic amyloidosis involving the interstitial tissues of the heart, the liver, the spleen, the adrenal glands and the uterus, as well as small and medium-sized arteries of these organs, together with those of the kidneys, the ovaries, the uterine tubes, the gastrointestinal tract, the lungs, the pancreas and the bone marrow; ischemic infarction of the left frontal lobe of the cerebrum, together with the basal ganglions on the left, secondary to arteriosclerotic thrombosis of the left midcerebral artery; focal bronchopneumonia; double right kidney and ureter; acute esophagitis, esophagomalacia and gastromalacia; multiple renal infarcts, bilateral; multiple small areas of pulmonary infarction.

COMMENT

This case fulfils the most important criteria of primary systemic amyloidosis in that there is no evidence of any preexisting condition recognized as capable of bringng about the formation of amyloid. The irregular and inconstant metachromatic staining with crystal violet is in keeping with observations of others; the subendocardial and myocardial deposits of amyloid in no way differ from those described in

many previous instances and reviewed by Lindsay.[7] The appearance of extensive deposition of amyloid in the liver, the spleen and the adrenal glands is less characteristic. Of the 22 cases reviewed by Koletsky and Stecher [3c] as instances of primary systemic amyloidosis, involvement of the spleen was observed in 2, adrenal involvement in 2 and hepatic deposition of amyloid, except for that found within the blood vessels, in none. The group of 16 cases reviewed by Lindsay and Knorp [3d] included 8 in which the hepatic parenchyma, 7 in which the spleen and 6 in which the adrenal glands were involved. This overlapping of distribution, involving sites characteristic of secondary amyloidosis, has been previously commented on.[8] Conversely, instances of amyloidosis of the secondary type have been reported in which the major sites of involvement were in the tissues of mesodermal origin, the secondary distribution simulating that seen in primary systemic amyloidosis.[10]

Pericellular deposits of amyloid occurring in adipose tissue have been described by Peters,[11] Iverson and Morrison [5] and Pearson and co-workers.[12] Peters considered such deposits to be the earliest form in which amyloid appeared. No evidence of such distribution has been found in adipose tissue in this case. In the myocardium, however, thin amyloid shells were present about the muscle fibers in the areas of least marked involvement (fig. 2 *B*). Larsen [13] concluded, from his study of serial sections, that amyloid is primarily a pericapillary deposit spreading into the interstitial tissue. In the case which he reported, there was no involvement of the coronary arteries, and he felt that the deposition of amyloid depended on changes of the permeability of the venous endothelium, since he often found amyloid in the myocardial venules. In our case, this distribution is reversed, and we find extensive amyloidosis of the coronary arteries with only minimal involvement of the cardiac veins. This question of the sites of the earliest or the most prominent deposition of amyloid is of some importance, for the answer has a bearing on the problem of the origin of the substance. Peters [11] cited German and Dutch authors of the early decades of this century, who are said to have described epicellular deposits of amyloid without having questioned the older doctrine that the material was formed as a transudate. He pointed out that deposition of amyloid occurred in the walls of arteries and referred to the difficulty of accounting for

9. Reiman and others.[3b] Koletsky and Stecher.[3c] Orloff and Felder.[8a] Golden.[4c] Iverson and Morrison.[5]

10. (*a*) Spain, D. M., and Barrett, R. C.: Arch. Path. **38**:203, 1944. (*b*) Budd, J. W.: Am. J. Path. **10**:299, 1934. (*c*) Lindsay and Knorp.[3d]

11. Peters, J. T.: Arch. Path. **35**:832, 1943.

12. Pearson, B.; Rice, M. M., and Dickens, K. L.: Arch. Path. **32**:1, 1941.

13. Larsen, R. M.: Am. J. Path. **6**:147, 1930.

a transudate in such an area in support of his contention that amyloid arises at the surfaces of cells. Larsen [13] noted medial amyloidosis of the aorta, the pulmonary vessels and the inferior vena cava, but he explained these deposits as related to the vasa vasorum, in accordance with his belief that amyloid was deposited from the intercellular fluid ("tissue lymph") as a result of altered venous permeability.

Mallory [14] held that amyloid was formed by perverted fibroblastic activity and that its being deposited immediately beneath vascular or sinusoidal endothelium was related to the fact that there were fibroblasts in the vicinity. Warren [15] supported and elaborated this contention, stating in connection with his report of a case of generalized muscular amyloidosis that the condition represented a widespread perversion of function of connective tissue elements of muscular structures of the body, involving smooth, striated and cardiac muscle. In his opinion there was no doubt that this was the mode of origin, for in many instances amyloid was found at a considerable distance from blood vessels.

It is manifestly impossible to draw conclusions regarding the early localization of amyloid from massive deposits such as are seen in the liver, the spleen and the adrenal glands in this case, as well as the relatively heavy accumulations in some portions of the myocardium and beneath the endocardium. The pericellular amyloid rings that we and others have seen are in keeping with the hypothesis that amyloid is produced by local cellular activity and tend to support the beliefs of Peters,[11] Mallory [14] and Warren [15] rather than the theories of those [16] who hold that amyloid is an abnormal protein precipitated from the body fluids.

In our case the chief feature of interest is the association of widespread amyloidosis of the cardiovascular system with massive deposits of amyloid in the liver, the spleen and the adrenal glands. This reemphasizes the fact that distribution alone does not differentiate primary from secondary systemic amyloidosis.

Of incidental interest is the autolysis of the fundus of the stomach and the lower part of the esophagus. This may be associated with the intracranial lesion.[17]

Many systems may be involved in primary amyloidosis, but they are seldom involved to a uniform extent. The pathologic change is frequently overshadowing in, and the symptom complex primarily

14. Mallory, F. B.: Principles of Pathologic Histology, Philadelphia, W. B. Saunders Company, 1914 (reprinted 1925).

15. Warren, S.: Am. J. Path. **6**:161, 1930.

16. Perla and Gross.[3a] Larsen.[13]

17. Moore, R. A.: Textbook of Pathology, Philadelphia, W. B. Saunders Company, 1945.

referable to, the cardiovascular system, the gastrointestinal tract, the lungs, the genitourinary system, etc. In the 50 cases previously reported an extensive involvement of the cardiovascular system has been common. In 24 cases there were clinical evidences of heart failure prior to death. In 21 of 48 fatal cases heart failure was indicated as a cause of death. Some degree of amyloid infiltration of the heart was found in 43 of 48 cases studied at autopsy.

Lindsay,[7] in his discussion of the heart involved in primary amyloidosis, lists the following mechanisms leading to heart failure:

1. Involvement of the pulmonary vessels and alveolar walls leading to chronic cor pulmonale (dilatation of the heart).

2. Amyloidosis of the cardiac vessels leading to coronary insufficiency or infarction.

3. Interstitial amyloid infiltration of the myocardium with or without secondary degeneration of muscle fibers.

4. Pericardial or endocardial deposition of amyloid.

5. Amyloid involvement of cardiac valves, with stenosis or insufficiency.

6. Combinations of the aforenamed mechanisms.

It would be expected that electrocardiographic findings would be variable depending on which of the aforementioned factors were dominant in the production of the heart failure. Diffuse infiltration of the myocardium would interfere with the strength of contraction and the flow of current through the heart walls. Detailed electrocardiographic reports are available in only 13 recorded cases. In 9 of these, heart failure was listed as a cause of death. Low voltage of the QRS complexes was the prominent feature in 7 cases and flattening or inversion of T waves in 2 cases.

In this case the cerebrovascular lesions were found to be due to thrombosis of arteriosclerotic vessels and were not related to the amyloid disease. The clinical picture was primarily that of cardiac failure of obscure cause, correlated at autopsy with amyloidosis of the myocardium. The abdominal enlargement with the grossly enlarged, hard liver and the globular-shaped heart led to consideration of mediastinopericarditis with pseudocirrhosis of the liver (Pick), although there was no evidence of constricton or fixation of the heart. The electrocardiogram showed low voltage and flattening of the T waves; these were the changes most prominent in previously reported cases of cardiac amyloidosis in which such studies were made.

The beefy red tongue and the cutaneous changes that were noted at the wrists and ankles of this patient were assumed clinically to be due to a vitamin deficiency, since there had been a poor nutritional

intake for a long period prior to hospitalization. Credence was lent to this view because of the definite and rapid improvement which occurred in these lesions under intensive vitamin therapy. Unfortunately, no sections were taken from these sites for microscopic study, since no gross abnormality could be noted at the time of autopsy and the nature of the general systemic disease was not then suspected. Amyloid involvement of the skin and the tongue has not been uncommon in the previously reported cases; whether there was such involvement in this patient and whether that involvement contributed to the clinical findings must remain a question.

It appears that this case further substantiates the observation of Lindsay [18] that primary systemic amyloidosis should be considered in the differential diagnosis of cardiovascular disease whenever the cause is obscure and the symptom complex and the clinical findings bizarre.

SUMMARY

Primary systemic amyloidosis is becoming more frequently recognized as a clinical entity. Its causation will presumably remain a mystery until the disputed chemical nature of amyloid is resolved. The literature is briefly reviewed, and an additional case is reported. Primary amyloidosis of the cardiovascular system should be considered in the presence of bizarre symptoms and findings of obscure cause, especially when the electrocardiogram indicates diffuse myocardial involvement.

18. Lindsay and Knorp.[8d] Budd.[10b]

MECKEL'S DIVERTICULUM WITH ABERRANT PANCREATIC TISSUE AND A POLYP CONTAINING GASTRIC GLANDS

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AND
H. A. TELOH, M.D.
CHICAGO

OF THE pathologic conditions due to congenital diverticulum of the small intestine, those associated with aberrant or heterotopic tissue are of the greatest interest. The occurrence of gastric mucosa, pancreatic tissue with or without islets of Langerhans, duodenal mucosa or colic mucosa is well known and has been frequently reported. In 1934 Hunt and Bonestiel [1] gave an excellent review of the occurrence of aberrant pancreatic tissue in the gastrointestinal tract including Meckel's diverticulum. However, the occurrence of a polyp in Meckel's diverticulum is rare, and only sporadic reports of it are found in the literature. The case reported now is considered of pathologic interest.

REPORT OF A CASE

A 51 year old white man was first admitted to Passavant Memorial Hospital, July 14, 1948, for bleeding gums, easy bruising, and numerous spots on his hands, arms and legs, present for three months. He had also noticed easy fatigability on exertion for a similar period. He had pneumonia in 1933 and underwent left herniorrhaphy in 1928.

The patient was well developed and well nourished. The temperature was 101 F; the pulse rate, 80; the respiratory rate, 20, and the blood pressure, 120 systolic and 70 diastolic. There were petechiae on the pale blue boggy buccal mucosa, an ulcerated area on the upper left gum and blood oozing from around the base of the lower left canine tooth. The lower extremities were covered with small red macular lesions and ecchymoses.

The red blood cell count was 2,540,000; the hemoglobin content, 10 Gm. The white blood cell count was 2,800, with 3 band cells, 21 segmented cells, 1 eosinophilic granulocyte, 71 lymphocytes, 2 myelocytes, 1 metamyelocyte and 1 monocyte. The specific gravity of the urine was 1.017; p_H, 6.0; albumin, a trace; sugar, none; red blood cells, 50 to 60; leukocytes, 20 to 30. The thrombocyte count was 7,620; the prothrombin time, 101.7 per cent. The blood urea nitrogen was 12.4 mg.; blood sugar, 108 mg.; blood ascorbic acid, 1.42 mg.; serum total protein 6.04 Gm.; albumin, 3.67 Gm.; globulin, 2.37 Gm., per hundred cubic centimeters.

From the Department of Pathology of Northwestern University Medical School and Passavant Memorial Hospital.

1. Hunt, V. C., and Bonestiel, H. T. S.: Arch. Surg. **28**:425, 1934.

A diagnosis of primary aplastic anemia was made.

On July 25, 1948 the patient was readmitted on account of marked pyuria. Cystoscopic and retrograde pyelographic examination led to a diagnosis of bilateral hydronephrosis and bilateral ureteral stricture with an enlarged prostatic bar. Postoperatively, there was profuse bleeding from the urinary tract. After five days this was controlled with blood transfusions.

The patient was readmitted on two subsequent occasions for blood transfusions. The blood picture remained unchanged.

Seven days after the date of the patient's fourth discharge, diarrhea developed, with four black stools during the day. Seven similar stools occurred on the following day.

The patient reentered the hospital on November 19. He was pale and listless. The oral temperature was 99 F.; the pulse rate, 112; the respiratory rate, 20, and the blood pressure, 104 systolic and 60 diastolic. Clotted blood was seen about the upper molar and lower incisor teeth, dried blood on the dorsum of the tongue and a small amount of blood with ecchymoses in the oropharynx. There were multiple petechiae and ecchymoses of the face. The abdomen was soft and nontender, and no organs or masses were palpable. Otherwise the examination disclosed nothing of importance.

The red blood cell count was 1,520,000; the hemoglobin content, 4.0 Gm. The white cell count was 1,550. No thrombocytes were seen. The specific gravity of the urine was 1.018; p_H, 6.0; albumin, 1 plus; sugar, none. The stool was black to dark red and gave a strongly positive reaction to the guaiac test. The patient received multiple massive transfusions over a period of twenty-four days, but severe intestinal hemorrhage continued. Surgical intervention was considered, but at no time was the patient considered to be in good enough condition to risk operation. He died twenty-four days after admission, of a massive hemorrhage of the rectum.

The anatomic diagnoses were: diverticulum of the small intestine with ulceration and acute and chronic inflammation; pedunculated polyp of the small intestine with aberrant gastric mucosa; aberrant pancreatic tissue in the diverticulum of the small intestine; hypoplasia of the bone marrow, marked; subdural hematoma, bilateral; intracerebral hemorrhages, multiple; acute passive hyperemia of the lungs, liver, spleen and kidneys; bilateral hydronephrosis and hydroureter, severe; chronic cystitis; hydrohemopericardium; chronic cholecystitis; cholelithiasis; generalized arteriosclerosis, slight; emphysema of the lungs, moderate; atelectasis of the lungs; chronic focal esophagitis; fibrosis of the testicles.

Sixty centimeters proximal to the ileocecal valve Meckel's diverticulum was encountered on the antimesenteric border of the ileum. This diverticulum measured 8 cm. in length and 3 cm. in diameter. The external surface was mottled gray to purple. On section the wall was thick and indurated and the mucous membrane deep purple to black. There were multiple areas of superficial ulceration of the mucous membrane. At the opening of the diverticulum there was a soft, friable polyp, 2.5 cm. in length and 0.8 cm. in maximum diameter. The surface was deep red to black and hemorrhagic in appearance. The intestinal contents proximal to this point were normal. Distally the contents were black and putty-like. There were numerous areas of ecchymoses scattered throughout the entire intestinal tract.

In microscopic sections through the diverticulum, the layers normally found in the small intestine were all present. Focally the muscularis was greatly attenuated or absent. There were marked diffuse infiltration of all layers with

acute and chronic inflammatory cells and diffuse interstitial edema, fibrosis and numerous focal areas of hemorrhage. The interstitial hemorrhage was most marked in the submucosa. The mucosa was partly autolyzed, and there were multiple focal areas of superficial ulceration. The base of the ulcerated areas was formed of necrotic debris, a thick layer of fibrin and polymorphonuclear leukocytes. In sections of the diverticulum there were small nodular areas in

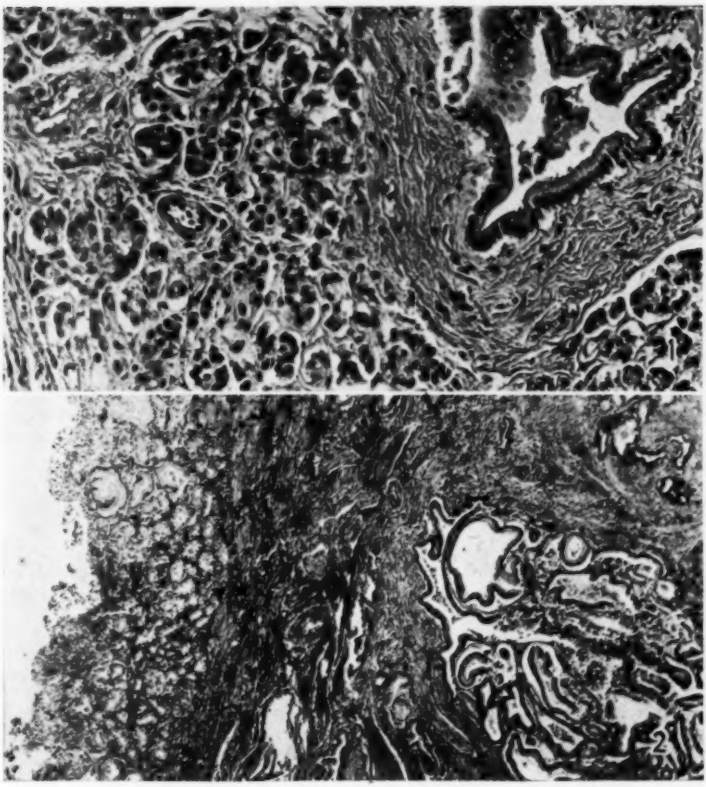

Fig. 1.—Pancreatic tissue in the submucosa of the diverticulum. × 90.
Fig. 2.—Gastric mucosa at the tip of the polyp and dilated, distorted gastric glands in the stromal tissue. × 90.

the submucosa, composed of pancreatic tissue; the largest of these measured 4 mm. in maximum diameter. There were no islets of Langerhans.

The section of the polyp consisted of an overlying mucosa and a dense fibrous stroma containing numerous glandular structures. The mucosa consisted throughout most of the surface of an intestinal type of glandular epithelium which was continuous with that of the small intestine and diverticulum. In this

mucosa there was a marked chronic inflammatory reaction with focal areas of interstitial hemorrhage. Around the tip of the polyp the mucosa was composed of typical gastric glands and also contained numerous chronic inflammatory cells and areas of hemorrhage. The gastric glands extended into the stroma for a considerable distance to form small and large, irregularly shaped and occasionally dilated glandular structures. The stroma consisted of a dense fibrous connective tissue with numerous focal areas of hemorrhage and marked diffuse infiltration with chronic inflammatory cells.

COMMENT

Meckel's diverticulum occurs in approximately 2 per cent of persons coming to autopsy. It occurs as a finger-like projection from the antimesenteric border of the ileum from 25 to 100 cm. proximal to the ileocecal junction. It may vary considerably in size, from a small flattened pouch of the ileal wall to a large tubular structure 10 cm. or more in length. Its diameter usually approximates that of the ileum.

Embryologically it represents a failure of obliteration of the fetal omphalomesenteric duct or vitellointestinal duct which forms the connection between the primitive alimentary canal and the yolk sac. Normally this becomes obliterated at the seventh week of fetal life. The presence of polyps in this region is unexplained. The fact that these polyps are frequently composed wholly or partly of heterotopic tissue would seem to exclude their fortuitous occurrence in Meckel's diverticulum.

According to the literature, there are 14 recorded cases of Meckel's diverticulum with a polyp. Many of these were incompletely studied pathologically and are necessarily discarded. Hertzler and Gibson [2] reported the case of a 19 year old boy with an intussusception associated with invagination of Meckel's diverticulum. The diverticulum contained a small papillary lesion composed of Brunner's glands. In a review of cases they enumerated 8 previously recorded cases of polyps of Meckel's diverticulum. In 7 of these the structures were not studied histologically. In the eighth, that of Mathieu and Davioud, the specimen consisted of a small polyp composed of pancreatic tissue. Schullinger and Stout [3] reported a case of massive hemorrhage of the bowel associated with Meckel's diverticulum. The diverticulum contained a small adenoma composed of gastric and duodenal glands. In a study of the literature they failed to find a similar example of gastric and duodenal gland adenoma of a diverticulum, and cited 3 previously reported cases of polyps. That of Lecene was an instance of Meckel's diverticulum with an adenoma at the apex. This, however, was covered by mucous glands resembling normal intestinal mucosa. The case of

2. Hertzler, A. E., and Gibson, E. T.: Am. J. M. Sc. **146**:364, 1913.
3. Schullinger, R. N., and Stout, A. P.: Arch Surg. **28**:440, 1934.

Bize was one of Meckel's diverticulum with a nodule at the apex which contained pancreatic glands with some portions having an adenomatous appearance consisting of large branching columnar cell–lined ducts. The third case was that of Hertzler and Gibson.

Starling [4] reported the occurrence of severe melena associated with a polyp. A "simple papilloma" was found a few inches beyond a diverticulum. No histologic examination is recorded. Bowen [5] summarized the previous literature and reported the case of a 14 year old boy with an intussusception. At operation Meckel's diverticulum with inversion was found in the affected bowel. It contained a polyp measuring 2 by 2 by 1.5 cm., also peptic glands.

In summary, of the cases found in the available literature, the following have been adequately studied histologically and are acceptable.

1. Mathieu and Davioud—polyp composed of pancreatic tissue.

2. Lecene—adenomatous polyp formed of normal intestinal mucosa.

3. Bize—nodule composed of pancreatic glands and large branching columnar cell–lined ducts.

4. Hertzler and Gibson—papillary lesion composed of Brunner's glands.

5. Schullinger and Stout—adenoma composed of gastric glands and Brunner's glands.

6. Bowen—polyp containing peptic glands.

Aberrant Pancreatic Tissue.—Aberrant or accessory pancreatic tissue was first described by Klob in 1859. The first recorded case of aberrant pancreatic tissue in Meckel's diverticulum was that of Zenker in 1861. It is an infrequent anomaly. Hunt and Bonestiel,[1] in a review of 186 cases of aberrant pancreatic tissue, found 13 cases involving Meckel's diverticulum. In 1940 Faust and Mudgett [6] reviewed 370 cases of aberrant pancreas, 21 of which were examples of occurrence in Meckel's diverticulum.

Zenker expressed the belief that it is due to an additional bud from the foregut which has developed into a simple independent glandular mass and that this, as the foregut elongates, may be carried a great distance from its point of origin. In support of his theory is the fact that aberrant pancreatic tissue may be found along the entire gastrointestinal tract from the stomach to the terminal portions of the ileum, as well as in the gallbladder, splenic capsule, umbilical fistula, omentum and mesentery.

4. Starling, H. J.: Guy's Hosp. Rep. **85**:207, 1935.

5. Bowen, F. H.: J. M. A. Georgia **30**:390, 1941.

6. Faust, D. B., and Mudgett, C. S.: Ann. Int. Med. **14**:717, 1940.

Various other theories have been suggested to account for the presence of gastric mucosa and pancreatic tissue in ectopic foci. Displacement of tissue during embryonic development may be due to the formation of additional anlage, with subsequent displacement along the gastrointestinal tract (Zenker's original theory), or it may be due to a transposition of tissue from the original site as a result of adhesions, either inflammatory or noninflammatory. Other authorities believe that it is due to a metaplasia of tissue during either fetal or adult life. The frequent presence of a marked inflammatory reaction would tend to support this theory. The atavistic theory supposes a reversion to a more primitive phylogenetic type as exemplified by certain lower animals and fishes. In these species one finds pancreatic tissue in the muscular coats of the intestinal tract, in the peritoneum and scattered diffusely through the intestinal tract. The theories of origin of heterotopic tissue are reviewed by Troll.[7]

Histologically, the aberrant tissue is identical with normal pancreatic parenchyma in structure and arrangement. Islets of Langerhans may or may not be present.

SUMMARY

A report of a case of massive intestinal hemorrhage complicating thrombopenia and primary refractory anemia is presented. At autopsy Meckel's diverticulum was found associated with a glandular polyp. The diverticulum contained aberrant pancreatic tissue; the polyp contained typical gastric mucosa and dilated, distorted gastric glands.

A review of the literature is presented and the acceptable reports of Meckel's diverticulum associated with a polyp are summarized.

The occurrence and the theories of origin of aberrant pancreatic tissue are briefly reviewed.

7. Troll, M. M.: Arch. Path. **38**:375, 1944.

Notes and News

Research Fellowships.—The American College of Physicians announces that a limited number of fellowships in medicine will be available from July 1, 1950 to June 30, 1951. These fellowships are designed to provide an opportunity for research training either in the basic medical sciences or in the application of these sciences to clinical investigation. They are for the benefit of physicians who are in the early stages of their preparation for a teaching and investigative career in internal medicine. Assurance must be provided that the applicant will be acceptable in the laboratory or the clinic of his choice and that he will be provided with the facilities necessary for the proper pursuit of his work. The stipend will be from $2,200 to $3,200.

Application forms will be supplied on request to The American College of Physicians, 4200 Pine Street, Philadelphia 4, and must be submitted in duplicate not later than October 1, 1949. Announcement of awards will be made in November 1949.

Announcements

THE AMERICAN CANCER SOCIETY FELLOWSHIPS IN EXFOLIATIVE CYTOLOGY

Regulations for 1948-1949

At the American Cancer Society's symposium on exfoliative cytologic diagnostic technics held in Boston in April 1948, it was the opinion of the invited delegates that facilities were urgently need for the training of qualified pathologists and clinicians in teaching positions at approved institutions providing residency training in pathology as well as facilities for training technicians.

At the same time the delegates recommended that the American Cancer Society proceed to engage actively the interest and support of institutions and laboratories in setting up such training programs where the best training facilities appeared to be available. Thus inquiries were sent to fourteen laboratories over the country, and on the basis of their interest and the monies available, grants will be made for fellowship training in ten laboratories to support twenty-three fellows.

PURPOSE OF TRAINING

To provide training in the diagnostic technics of exfoliative cytology for qualified pathologists. It is anticipated that trainees will not assume the role of teachers until sufficient personal experience and competence have been acquired.

AWARDS

Fellowships will be awarded by institutions or laboratories designated by the Society to applicants on the basis of their past training and their intention to teach in their own laboratories diagnostic technics in exfoliative cytology to interested pathologists, clinicians and technicians.

ELIGIBILITY OF APPLICANT

The applicant for a Fellowship in Exfoliative Cytology of the American Cancer Society shall:

1. Be a graduate of a Class A Medical School of the United States, its Territories or Canada
2. Be a citizen of the United States

3. Be not over 50 years of age on the next birthday following commencement of fellowship tenure
4. Have completed two years of post-graduate training in pathology
5. Conform in other respects to requirements of the institution to which he applies.

TERM OF FELLOWSHIP

Each fellowship will be awarded for a period of four months. Fellowship training may commence at any time. The fellowship is not subject to renewal.

STIPEND

The stipends shall be paid in two sums: the first, a grant to the laboratory for tuition, overhead, other expenses as outlined to the Society; the second, to the trainee to partially cover his board, room and incidentals for the period of training. The latter sum shall amount to $140 per month and shall be paid direct to the fellow monthly in advance to avoid payment of income tax for services rendered to the hospital.

APPLICATION

The individual applicant for a Fellowship in Exfoliative Cytology shall apply directly to the institution where a fellowship is available. In no instance shall application be made directly to the American Cancer Society.

FELLOWSHIPS AVAILABLE

Laboratory	Number	Director
Cornell University Medical College, New York	6	George N. Papanicolaou, M.D.
Jefferson Hospital, Philadelphia	6	Lewis C. Scheffey, M.D.
University of Oregon Medical School, Portland, Ore.	2	Warren C. Hunter, M.D.
University of California Hospital, San Francisco	2	Herbert F. Traut, M.D.
Michael Reese Hospital, Chicago	2	Otto Saphir, M.D.
Hartford Hospital, Hartford, Conn.	2	Ralph E. Kendall, M.D.
New York Post-Graduate Hospital, New York	1	Locke L. Mackenzie, M.D.
Free Hospital for Women, Brookline, Mass.	1	Arthur T. Hertig, M.D.
Mayo Clinic, Rochester, Minn.	1	John R. McDonald, M.D.

Books Received

CLINICAL ASPECTS AND TREATMENT OF SURGICAL INFECTIONS. By Frank Lamont Meleney, M.D., associate professor of clinical surgery, College of Physicians and Surgeons; associate visiting surgeon, Presbyterian Hospital, New York. With a foreword by Allen O. Whipple, M.D. Pp. 840, with 289 illustrations. Price $12. Philadelphia and London: W. B. Saunders Company, 1949.

This book consists of eighteen chapters. It presents a documented history of the development of the treatment of surgical infections and emphasizes the fundamental surgical principles as well as the newer therapeutics, such as chemotherapy and the use of antibiotics. It is well illustrated with drawings, photographs and case reports. The various areas included in general surgery are discussed and also the surgical specialties. The author has made great contributions to the field of surgical bacteriology and is especially well qualified to produce a text on this subject. In addition, Dr. Meleney has enlisted the help of a number of able scientists, whose investigations and experience enhance the value of this work. The presentation includes physiologic, bacteriologic and pathologic aspects of the lesions as well as important factors of both diagnosis and treatment. Because of the excellent manner in which the material is presented and the scientific background of the writers, this book will fill the long-standing need of a source of information and will occupy an unusual position. It is an excellent book for the use of both medical students and practicing surgeons.

BLOOD TRANSFUSION. By Elmer L. DeGowin, M.D., associate professor of internal medicine, State University of Iowa, and director of the blood transfusion service of the University Hospitals; Robert C. Hardin, M.D., assistant professor of internal medicine, State University of Iowa, and John B. Alsever, M.D., senior surgeon, United States Public Health Service, and chief, Professional Standards, Hospital Division, United States Public Health Service. Pp. 587, with 200 diagrammatic drawings. Price $9. Philadelphia and London: W. B. Saunders Company, 1949.

It is a great pleasure to find a book in which authors and publishers have succeeded not only in satisfying the intelligence of the reader but in gratifying his artistic sense as well. It is attractively bound, is printed on good paper and is interestingly illustrated. The text includes a remarkably complete discussion of the ways in which whole blood and its constituent parts are used in the treatment of human disease. The book begins with a brief historical chapter and then proceeds immediately to a description of the therapeutic value of blood and of the blood derivatives and plasma substitutes that are available for intravenous, intramuscular and topical use. The cause and the treatment of shock are described. This is followed by an excellent section devoted to a discussion of ABO, MN, P and RhHr blood groups. The inheritance of blood groups, the occurrence and the immunizing capacity of the various antigens and the clinical aspects of immunization are handled clearly and completely.

The second section is devoted largely to technics and should prove extremely valuable to any one concerned with any part of a transfusion service. It begins with detailed, well illustrated descriptions of technics for determining antigens and antibodies and describes the other steps necessary in preparing for a trans-

fusion or in studying various pathologic states. It then gives methods for measuring the survival time of erythrocytes, for estimating bilirubin and urobilinogen in blood and excreta and for determining specific gravity and total blood volume. Chapters are devoted to donors (their selection, the technic of drawing blood, the possible complications) and to recipients (the methods by which transfusions can be given and the cause, diagnosis, prevention and treatment of transfusion reactions). Methods of storing blood and transporting it are described. The merits and drawbacks of plasma and the methods by which it can be prepared and fractioned are followed by the use of red cell suspensions, blood derivatives and plasma substitutes. The practical experience of the authors in running blood banks makes the chapters on blood banks and on community, regional and state blood services especially valuable. The book closes with descriptions of the apparatus for giving blood transfusions and for preparing fluids for parenteral therapy.

This book presents a more comprehensive survey of the theoretic and practical aspects of blood as a therapeutic agent than has ever before been available, and it should answer a growing need arising as a result of the increasing appreciation of the therapeutic value of blood and its derivatives.

CURRENT THERAPY, 1949; LATEST APPROVED METHODS OF TREATMENT FOR THE PRACTICING PHYSICIAN. Howard F. Conn, M.D., editor. Consulting editors: M. Edward Davis, Vincent J. Derbes, Garfield G. Dunsan, Hugh J. Jewett, William J. Kerr, Perrin H. Long, H. Houston Merrett, Paul A. O'Leary, Walter L. Palmer, Hobart A. Reimann, Cyrus C. Sturgis, Robert H. Williams. Pp. 672. Price $10. Philadelphia and London: W. B. Saunders Company, 1949.

ATLAS OF ORAL AND FACIAL LESIONS AND COLOR FILM LIBRARY. By Ralph Howard Brodsky, D.M.D., consulting oral surgeon of the Department of Hospitals, New York; lecturer in stomatology, Graduate School of Medicine, New York University; associate dentist to the Mt. Sinai Hospital, New York. Pp. 129, with 100 color slides. Price $80. Baltimore: Williams & Wilkins Company, 1949.

This is a teaching unit consisting of a cloth-bound text of 127 pages and a case with 100 color slides bearing numbers that correspond to numbers in the text. In the book is a description of each slide with diagrams indicating the particular condition each slide illustrates. The book and the slides should be of value in teaching and in diagnosis.

AN ATLAS OF BONE-MARROW PATHOLOGY. By M. C. G. Israëls, M.Sc., M.D., M.R.C.P., lecturer and deputy director of the department of haematology of the University and Royal Infirmary, Manchester. Illustrations by D. Davison, medical artist to the University of Manchester. Price $6.50. Pp. 79, with 3 figures and 12 color plates. New York: Grune & Stratton, 1948.

This little volume is designed to induce pathologists to examine the bone marrow and to tell the clinician the results of such examinations. The book is in two sections. The first deals with technic and the identification of the various cell types. The second part describes the marrow patterns found in different diseases. The clinician will welcome the comprehensive tabulation of the typical findings in the various blood disorders. The pathologist may consult the table which lists the diagnoses compatible with different marrow pictures. The first seven color plates depict the normal and the pathologic cells. The last five plates contain twenty characteristic marrow patterns. The illustrations are excellent. This atlas can be highly recommended to any student of blood diseases.

OBSERVATIONS ON THE PATHOLOGY OF HYDROCEPHALUS. By Dorothy S. Russell. Medical Research Council Special Report Series no. 265. Price 6 shillings. Pp. 138, with 90 illustrations. London: His Majesty's Stationery Office, 1949.

This report on hydrocephalus is a thorough and lucid discourse on the subject. Interest and information are added by critical reviews of other publications. The author in meeting Spiller's reproach "that actually observed lesions (of hydrocephalus) are much rarer than theories explanatory of the causes of hydrocephalus" presents a wealth of material which refutes the "ideopathic" origin of hydrocephalus. She systematically classifies her cases according to etiology. Maldevelopments are critically analyzed, and reasons for disagreeing with current theories are objectively presented. Inflammation is fully considered, the author commenting on the high incidence of Bacillus coli meningitis in the newborn. The entire problem of neonatal meningitis is considered at great length. Syphilis as a cause of hydrocephalus is rare in the author's experience. Numerous case histories, complete with postmortem findings and excellent illustrations, and an extensive bibliography are presented. Pathologists and neurologists will find it a useful text and reference source.

DIE PATHOLOGISCH-ANATOMISCHEN GRUNDLAGEN DER ALLERGIE. Von Doz. Dr. med. Wilhelm Eickhoff, Rheine, Westphalia. Pp. 95, with 40 illustrations. Stuttgart: Georg Thieme Verlag, 1948.

The purpose of this booklet is to introduce the beginner into the complex field of allergy by a presentation of the pathologic-anatomic features of the subject. The book is divided into two parts: experimental allergy and parallergy in the animal, and a short review of knowledge of human allergy as applied to immunization phenomena. There are many good photographs of the gross and histologic aspects of allergic phenomena. Unfortunately, no references to the literature are given. It is a well organized introduction to a difficult subject from a new point of view and should be of value to everybody interested in the theoretic basis of allergy.

EVALUATION OF CHEMOTHERAPEUTIC AGENTS. Edited by Colin M. MacLeod. Symposium held at the New York Academy of Medicine, March 25 and 26, 1948. Price $4. Pp. 205. New York: Columbia University Press, 1949.

The book contains the fourteen papers of the symposium. The authors are active investigators in chemotherapy. The papers deal instructively with general factors concerned in chemotherapy rather than with specific compounds for particular diseases.

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Subject entries are made for all articles. Author entries are made for original articles. Book Reviews are indexed under this heading in alphabetical order under the letter B.

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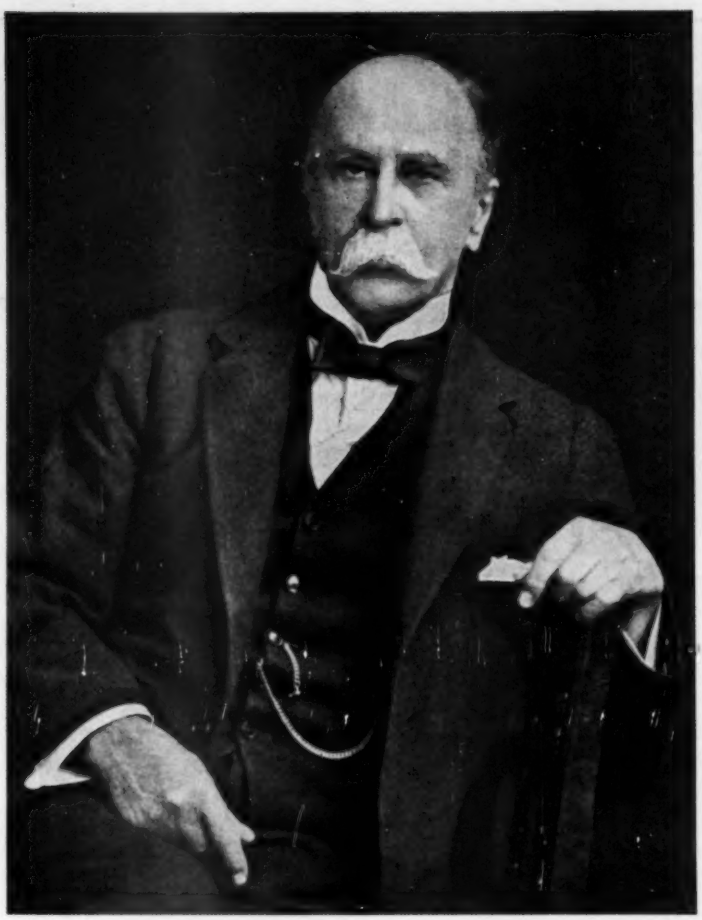

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